AD NUMBER

AD062764

LIMITATION CHANGES

TO:

Approved for public release; distribution is unlimited.

FROM:

**Distribution authorized to U.S. Gov't. agencies and their contractors;
Administrative/Operational Use; NOV 1954. Other requests shall be referred to Air Force Wright Air Development Center, Aero Medical Laboratory, Wright-Patterson AFB, OH 45433.**

AUTHORITY

afwal notice, 3 nov 1983

WADC TECHNICAL REPORT 54-344

THE TOXICITY OF CERTAIN LUBRICANTS, ENGINE OILS, AND CERTAIN OF THEIR CONSTITUENTS, WITH PARTICULAR REFERENCE TO THE PRODUCTS OF THEIR THERMAL DECOMPOSITION

JOSEPH F. TREON
FRANK P. CLEVELAND
JOHN CAPPEL

KETTERING LABORATORY
UNIVERSITY OF CINCINNATI

NOVEMBER 1954

WRIGHT AIR DEVELOPMENT CENTER

WADC TECHNICAL REPORT 54-344

THE TOXICITY OF CERTAIN LUBRICANTS, ENGINE OILS, AND CERTAIN OF THEIR CONSTITUENTS, WITH PARTICULAR REFERENCE TO THE PRODUCTS OF THEIR THERMAL DECOMPOSITION

JOSEPH F. TREON
FRANK P. CLEVELAND
JOHN CAPPEL

KETTERING LABORATORY
UNIVERSITY OF CINCINNATI

NOVEMBER 1954

AERO MEDICAL LABORATORY
CONTRACT No. AF 33(038)-26456
RDO No. 698-31

WRIGHT AIR DEVELOPMENT CENTER
AIR RESEARCH AND DEVELOPMENT COMMAND
UNITED STATES AIR FORCE
WRIGHT-PATTERSON AIR FORCE BASE, OHIO

Carpenter Litho & Prtg. Co., Springfield, O.
200 - 21 April 1955

FOREWORD

The research reported herein was performed in the Kettering Laboratory in the Department of Preventive Medicine and Industrial Health, College of Medicine, University of Cincinnati, Cincinnati, Ohio, under Contract No. 33(038)26456 with the Aero Medical Laboratory, Directorate of Research, Wright Air Development Center, Wright-Patterson Air Force Base, Ohio.

The work was carried out under RDO No. 698-31, "Toxicity of Developmental Air Force Materials", with George Kitzes, Ph.D., as contract monitor.

Actively engaged in the work were Joseph F. Treon, Ph.D., Frank P. Cleveland, M.D., John Cappel, B.S., Edwin E. Larson, M.S., Ralph W. Atchley, B.S., Frederic E. Shaffer, M.S., John P. Torbeck, B.S., Leo A. Hartman, B.S., James Gosney, B.S., Irving Wittow, B.S., Jose Gotay, B.S., Thomas Gahegan and George Neddermann.

The work was conducted and the report prepared under the general supervision of Francis F. Heyroth, M.D., Director of the Division of Toxicology in the Department of Preventive Medicine and Industrial Health, College of Medicine, University of Cincinnati, and was edited and approved by Robert A. Kehoe, M.D., Director of the Department.

ABSTRACT

Di-2-ethylhexyl sebacate, di-sec-amyl sebacate, di-2-ethylhexyl adipate, and formulations WS-2211 and PRL-3313, when given undiluted in a single oral dose to animals are either slightly toxic or practically non-toxic. The esters and formulations WS-2211 and PRL-3039 when maintained in contact with either the intact or abraded skin of rabbits for 24 hours are practically non-toxic. Repeated applications of large doses of WS-2211 upon the skin of rabbits resulted in irritation.

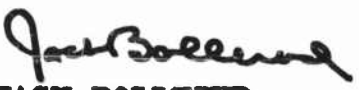
High concentrations of the mists of these substances were generally tolerated without untoward reactions.

The highest tolerable dosage and the lowest lethal dosage for one or more species of animals exposed to fog of the thermal decomposition products formed from these materials at 700°F have been established and are expressed in terms of milligrams of the substance delivered into the Inconel furnace per liter of air passing through it.

PUBLICATION REVIEW

This report has been reviewed and is approved.

FOR THE COMMANDER:



JACK BOLLERUD
Colonel, USAF (MC)
Chief, Aero Medical Laboratory
Directorate of Research

TABLE OF CONTENTS

SEQUENCE OF TABLES

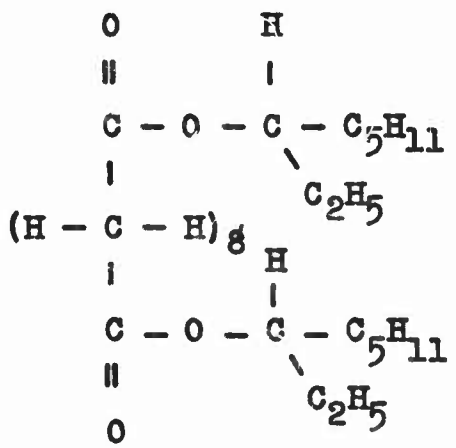
LIST OF ILLUSTRATIONS

INTRODUCTION

The purposes of the work reported herein were (1) to determine the immediate toxicity of di-2-ethylhexyl sebacate, di-sec-amyl sebacate, di-2-ethylhexyl adipate and formulations containing certain of these esters, namely, WS-2211 (both unused and used), PRL-3313 and PRL-3039, when given orally to rabbits and rats and when maintained upon the intact and abraded skin of rabbits for 24 hours; (2) to test the cumulative action of a series of small doses of WS-2211 (both before and after use in an airplane motor), when given orally to rabbits and rats or when maintained in contact with the skin of rabbits for 2 hours on each of 5 days per week over a period of several weeks; (3) to test the effects when the esters were inhaled as a vapor by animals of several species for 7 hours on 5 consecutive days; (4) to determine the toxic effects of mists of the three esters or of formulations WS-2211 and PRL-3313, when inhaled by animals of several species for 7 hours on each of 10 days over a period of 2 weeks; (5) to determine the immediate toxicity of the three esters and of formulations WS-2211 and PRL-3039, when inhaled as a fog formed by dropping them upon an Inconel surface maintained at an elevated temperature; and (6) to describe the histopathologic changes encountered in the tissues of animals exposed to these materials under the various experimental conditions.

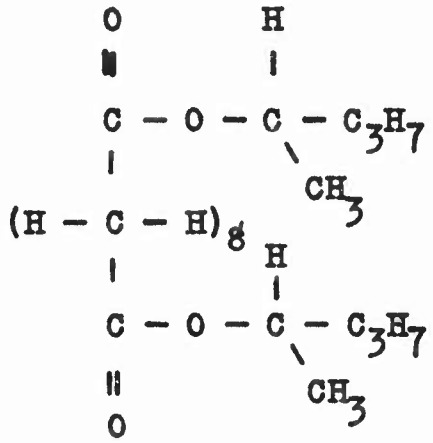
PROPERTIES OF MATERIALS

Di-2-ethylhexyl sebacate (Plexol 201, registered Trade Name of Rohm and Haas),



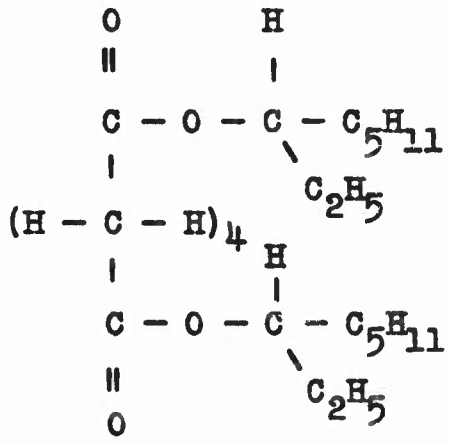
is a clear yellow fluid which has the molecular weight of 426.660.

Di-sec-amyl sebacate (Plexol 202, registered Trade Name of Rohm and Haas),



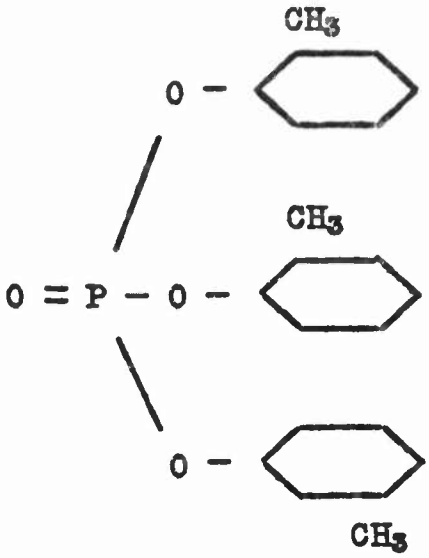
is a clear light straw-colored fluid, the molecular weight of which is 342.504. It has a flash point of 372°F and a fire point of 425°F.

Di-2-ethylhexyl adipate,

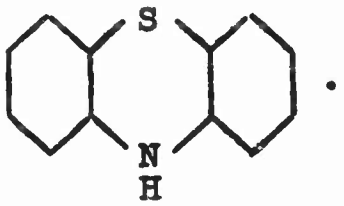


is a clear colorless liquid, the molecular weight of which is 370.556. Its specific gravity at 20°C is 0.925, its flash point, 400°F and its fire point, 450°F.

Formulation WS-2211 was prepared by the Standard Oil Development Company. It consisted principally of di-2-ethylhexyl sebacate, small amounts of a complex ester (ED-35), mixed tricresyl phosphates (Ohio Apex-Kronitex AA), one of which is shown below,



and a very small amount of phenothiazine,



The material employed for investigating the mists and fog formed by thermal decomposition was supplied to us by the Aero Medical Laboratory of Wright Air Development Center, which had received it as purchased from the Standard Oil Development Company. This turbo-jet lubricating oil, which fulfilled the military specifications of MIL-L-7808, was reported to have the specific gravity at 60/60°F of 0.929. It had not previously been used in a motor.

The Standard Oil Development Company also supplied a sample of unused WS-2211 (coded MRD-52-8). In addition this company also supplied a sample of WS-2211 (coded MRD-52-9) that had been used for 63 hours in a J-57 engine at the Pratt and Whitney Laboratory. The temperature of this engine, while running, was usually 600°F, and never below 250°F. These later samples were employed for the determination of the toxicity of the material when given orally to rabbits and rats and when maintained upon the skin of rabbits on one or more occasion. The Standard Oil Development Company, which sponsored this phase of the work, has granted permission for the inclusion of the data herein.

Formulation PRL-3313, which was a light red-brown fluid, consisted principally of di-2-ethylhexyl sebacate, with small amounts of Acryloid HF 25 and mixed tricresyl phosphates, and very small amounts of phenothiazine and Ortholeum 162 (dilauryl acid phosphate).

Formulation PRL-3039 consisted principally of di-sec-amyl sebacate and small amounts of di-2-ethylhexyl sebacate, Acryloid HF 25 and mixed tricresyl phosphates, and a very small amount of phenothiazine. This formulation is a red-brown fluid which has a specific gravity of 1.15 to 1.18 at 20/20°C, a flash point of 350°F and fire point of 375°F.

EXPERIMENTAL METHODS

Oral Administration of a Single Dose

A measured amount of one of the undiluted materials was introduced into the stomach of each of a series of healthy female rabbits from an hypodermic syringe by means of a rubber catheter (F20) passed through the esophagus. To make certain that the entire dose entered the stomach, the tube was flushed with 4 or 5 ml of water. In the case of rats, a measured amount of the undiluted material, contained in a syringe, was expelled through a blunt, 6 cm, 17-gauge hypodermic needle inserted into the esophagus. In this case the volume of material was measured by displacement from the syringe, the needle having been filled with the compound before it was introduced, and therefore the syringe was not flushed out with water. Animals were weighed daily until any losses sustained following the administration of one of the materials had been regained, after which they were weighed once per week until they were killed.

Oral Administration of a Series of Doses

In the determination of the effects of oral administration of multiple doses of WS-2211 (MRD-52-8 and MRD-52-9) to rabbits and rats, 10 and 25 per cent V/V solutions in peanut oil, respectively, were given in the manner indicated above on each of 5 days per week over a period of several weeks.

Application Upon the Skin of Rabbits

By means of the technique of Draize, Woodard and Calvery, the dosage of 9.4 ml of the undiluted product to be tested per kilogram of body weight was maintained for 24 hours in contact with the intact skin of each of 3 rabbits and with the abraded skin of each of 3 others. If fatalities resulted in either group, a corresponding experiment was carried out at a lower dosage (6.0 ml per kg). The hair was clipped from an area of skin 6 to 7 inches wide and completely encircling the trunk of each rabbit. The entire trunk was covered with a sleeve of dental dam which fitted tightly at the cephalad and caudad edges of the clipped area and loosely over the intervening area. During the 24-hour period of contact the rabbit was kept in stocks. The outermost layer of the skin of certain rabbits was abraded by scratching thin furrows 7.5 cm in length and a few millimeters apart with an hypodermic needle, prior to the application of the material being tested. At the end of 24 hours, the material was removed from the skin of the animals by washing with water and a sulfonated oil, "pH6".

Repetitive Applications Upon the Skin of Rabbits

In a second series of experiments, 4 groups of 3 rabbits were kept in stocks in the supine position, the hair having been previously clipped from the abdominal skin. In the case of 2 groups, the dosage of 5.0 ml of WS-2211 (either undiluted MRD-52-8 or MRD-52-9) was maintained in contact with the intact abdominal skin for 2 hours per day on each of 8 days over a period of 10 days. The other 2 groups were subjected in like manner to contact with the respective materials employed in the dosage of 2.0 ml per day on 5 days over a period of 10 weeks.

Volatilization of Undecomposed Vapor

A group of animals consisting of 2 male guinea pigs, 5 male mice, 3 female rabbits and 3 male rats was exposed to air nearly saturated with di-2-ethylhexyl adipate for 7 hours per day on each of 5 consecutive days in a 223-liter, steel cylindrical chamber (Figure 4). A similar group, except that the mice were females, was exposed to di-2-ethylhexyl sebacate for a corresponding period. In a corresponding

investigation of the effects of di-sec-amyl sebacate, 2 female guinea pigs, 5 male mice, 2 female rabbits, 3 male rats and a female cat were employed.

For the volatilization of any of these esters two flat-bottomed cylindrical bubbling towers (2.94" x 13") were employed in parallel. A fritted-glass thimble (0.94" x 3.75") was immersed in the liquid (600 ml) contained in each (Figure 1). These towers were kept in a bath of Nujol maintained at approximately 110°C. Air passed through the two bubbling towers at rates of 10.0 and 10.7 liters per minute, respectively, as measured by a rotameter in each line preceding the tower. The streams merged in a glass tube leading to a manifold immersed in a cooling bath before entering the chamber. Some condensation occurred in the effluent tube of the bubblers.

Aspiration of a Mist at Room Temperature

The mists of the esters and of formulation WS-2211 were generated by use of the glass injector shown in Figure 2 and described in detail in the publication "The Toxicity of Sulfuric Acid Mist" (Treon, Dutra, Cappel, Sigmon and Younker). In the formation of air-borne mists, a stream of dried compressed air at 1.27 to 3.47 atmospheres of pressure passed over an orifice at the end of a tube, the lower portion of which was immersed in an Erlenmeyer flask containing the material. The air then entered the chamber at rates within the range of 5.6 to 48 liters per minute at room temperature and atmospheric pressure. The larger droplets impinged on a mantle and returned to the Erlenmeyer flask. In a few instances (Table 7) uncontaminated supplementary air was passed into the chamber.

The mist of formulation PRL-3313 was generated by the use of a Lundegardh type of atomizer, a sketch of which is shown in Figure 3. A stream of compressed air (at a pressure of 2.5 atmospheres in one experiment and 1.3 atmospheres in a second experiment) passed from the gauge of a compressor through a silica gel for drying before entering the atomizer containing the oil in the outer mantle. The atomizer was composed essentially of a glass vessel into the bottom of which was inserted a brass tube (A), bearing at its end a stainless steel shaft (C), terminating in a removable brass jet surrounded by a stainless steel

nozzle (D). The tube was held in place in a glass 19/38 $ male joint by a lock-nut (B) and a rubber washer. The shaft was 0.64 cm in outside diameter, 0.24 cm in inside diameter and had at the top a hole 0.044 cm (brass jet) in diameter. The stainless steel nozzle was immersed in a layer of the liquid, which entered the space surrounding the jet through a hole 0.6 cm in diameter. On the removable upper portion (E) of the stainless steel nozzle was a brass disk (F) perforated by a 0.10 cm opening which widened out to a 45° angle. The air, in passing through the tube (A), sucked up the liquid surrounding the jet and carried it as a spray through the opening (0.32 cm) in the adjustable upper portion of the nozzle (E). The larger droplets of the spray were returned to the main portion of the liquid after striking the walls of the vessel, while the air carrying the fine mist passed through the side-arm (G) into the experimental chamber through an opening toward the rear of its top, where it was distributed by means of a fan mounted on the rear wall.

During the exposure of the animals to a mist, they were confined in a 223-liter chamber as shown in Figure 4.

Generation of a Fog From a Heated Inconel Surface

The equipment for the thermal generation of fogs from di-2-ethylhexyl adipate and from formulation WS-2211 (unused sample) is shown in Figure 4. A rotameter for measuring the flow of air, a furnace for volatilizing and decomposing the fluids, a semi-potentiometer, a glass cooling manifold, and the 223-liter chamber for exposing the animals are shown. In these experiments the smaller rotameter, to the right in the photograph (Figure 4), was not employed, and all of the air (31.8 liters per minute) that entered the chamber was passed through the furnace. The glass cooling coils were packed in the bath shown, and surrounded by crushed ice.

The air supplied to the furnace by two Gast Rotary Blast and Suction pumps was dried by passing through a series of six towers (see Figure 5 of the report of June 16, 1953) containing, respectively, (1) concentrated sulfuric acid, (2) air, (3) glass wool, (4) calcium chloride, (5) sodium hydroxide, calcium chloride and Drierite, and (6) activated charcoal.

The body of the furnace consisted of a 26-inch length of cold-drawn, seamless Inconel tubing, having an outside diameter of 1.5 inches and a wall thickness of 0.049 inch. Di-2-ethylhexyl sebacate was dropped from the bottle containing the material at constant pressure, through a graduated, air-cooled, glass orifice within an Inconel tube welded to the upper surface of the furnace near its midpoint. This tube had a length of 6 inches, an outside diameter of 0.675, and a wall thickness of 0.091 inch. Another piece of Inconel tubing 3 inches in length, and having an outside diameter of 0.438 inch and a wall thickness of 0.049 inch, was also welded to the upper portion of the furnace tube, to permit the insertion of an iron-constantan thermocouple in such a manner as to measure the temperature at the upper interior surface of the furnace. The distance from center to center of the two smaller tubes, which were parallel to one another and perpendicular to the furnace proper, was 1.75 inches.

The outside of the furnace was wrapped with two layers of molding mica. The middle 10-inch portion of the length of the furnace was electrically heated by means of 20 feet (9 wrappings to the inch) of A.W.G. Number 20 Tophet "C" wire, which has a resistance of 0.659 ohm per foot at 68°F. The wire was insulated by means of approximately 0.75 inch of diatomaceous earth and wrapped on the outside with asbestos cloth.

The temperature of the wall was regulated by a Variac and was measured by an iron-constantan thermocouple attached to a semi-potentiometer calibrated against a student potentiometer. The thermocouple was in contact with the upper part of the furnace about 1 inch downstream from the point at which the droplets of material fell upon its lower surface.

The animals were exposed to the thermal decomposition products of di-2-ethylhexyl sebacate, di-sec-amyl sebacate or formulation PRL-3039 in an 800-liter chamber (Figures 5 and 6). The fogs were generated in an 18-inch furnace (Figure 7) equipped with cooling coils made from Stainless Steel Carpenter Type 304 through which the decomposition products passed before entering the exposure chamber. The 9-inch portion of the furnace downstream from the entry of the liquid, and the 4-inch portion upstream, were electrically heated by means of 22.5 feet of Nichrome wire (0.032" diameter, 20 B & S), which had a resistance of 0.666

ohm per foot. The furnace was wrapped first with 2 layers of molding mica, then alternately with the Nichrome wire and asbestos cord and then insulated with asbestos tape and fiber glass. The rate of delivery was ascertained by observing the number of puffs of smoke per minute, the relation of the number of puffs per minute to the weight of liquid delivered per minute having been established previously. In each experiment, the total amount of material delivered was determined by weighing the reservoir before and after the experiment.

Sampling and Analysis of Contaminated Air

Mist from Unheated Ester. On each day, 1 or 2 samples of air were withdrawn from the chamber at the rates of 1 to 3 liters per minute for 60 minutes, through 2 thirteen-inch absorption towers (2.25" outside diameter) each containing 100 ml of 95 per cent ethyl alcohol. The sample passed through coarse fritted-glass discs (1.13" in diameter) immersed in the solvent and situated parallel to the bottom of the tower and about 0.2" above it.

In the case of either di-2-ethylhexyl sebacate or formulation PRL-3313, the total sample was evaporated to a few milliliters on a hot plate, and then transferred with alcohol to a 10 ml beaker. The contents were evaporated at a low temperature to a clear liquid, until its weight became constant. When the materials under investigation were di-sec-amyl sebacate, di-2-ethylhexyl adipate, WS-2211 or PRL-3313, the sample so collected was subjected to hydrolysis. When animals were being exposed to the vapor, rather than to the mist, of one of the three esters, the concentration was determined by hydrolysis and titration. One milliliter of 0.1 N alkali is equivalent to the following amounts of the different esters:

Di-2-ethylhexyl sebacate	21.33 mg
Di-2-ethylhexyl adipate	18.53 mg
Di-sec-amyl sebacate	17.13 mg

Three milliliters of 0.5 N alcoholic KOH were refluxed for 0.5 hour with a 25 ml aliquot of the sample. After the addition of 100 ml of water, the sample was titrated with 0.1 N aqueous hydrochloric acid employing phenolphthalein as an indicator. This method yielded satisfactory recoveries of known quantities of the

esters added to alcohol. The concentrations of mists of formulations WS-2211 and PRL-3313 were expressed in terms of di-2-ethylhexyl sebacate.

Products of Thermal Decomposition. Total aldehydes (expressed as formaldehyde), formaldehyde, carbon monoxide and undecomposed ester were determined in the fogs formed from each of the 3 esters and from formulations WS-2211 and PRL-3039. The amounts of total particulate matter in the fogs from all of the materials except WS-2211 were determined, as was the content of carbon dioxide in all except those that arose from di-2-ethylhexyl adipate and from WS-2211.

Carbon monoxide was determined by a slight modification of the method of Bland. The samples were collected by passing the fogs successively through a solution of lead acetate, concentrated sulfuric acid, silica gel impregnated with layers of a cobalt salt, magnesium perchlorate, a sodium hydrate-asbestos absorbent, and a second layer of magnesium perchlorate. The air then passed through a heated tube containing iodine pentoxide. The liberated iodine was collected in a bubbler containing an aqueous solution of potassium iodide (Figure 8) and titrated with 0.001 N arsenious acid.

Carbon dioxide was collected by passing air through 2 absorption towers (Figure 5) each containing 100 ml of 0.1 N aqueous sodium hydroxide. An aliquant was titrated with 0.1 N hydrochloric acid, employing phenolphthalein and methyl orange as indicators according to the method of Willard and Furman.

Total aldehydes were determined by the method of Goldman and Yagoda, which is based upon the ability of aldehydes to form a complex salt, sodium acid sulfite, that is stable in slightly acid and neutral solutions, but is decomposed to the aldehyde in an alkaline solution. These samples were collected in 2 absorption towers (Figure 5) each containing 100 ml of 1 per cent aqueous sodium acid sulfite. The sodium acid sulfite remaining in excess is destroyed by iodine in a neutral solution. The solution is then made alkaline and the liberated sulfite is titrated with standardized iodine. In the case of the fog formed from di-2-ethylhexyl adipate, an additional sample of air was passed through a similar absorption train containing the 1 per cent bisulfite in a mixture of equal volumes of water and 95 per cent ethyl alcohol.

Formaldehyde was determined by the chromotropic acid method of Bricker and Johnson in another portion of the bisulfite sample.

The total particulate matter was determined by washing the deposit collected in an electrostatic precipitator (Figure 5) with alcohol, and weighing the residue after evaporation of the solvent.

Each of the following concentrations of the various contaminants of the air of the chamber, expressed as p.p.m. by volume in air at 25°C and 745 mm of mercury, is equivalent to the concentration of 1 mg per liter.

Compound	1 mg/liter = p.p.m.
Carbon monoxide	891
Carbon dioxide	567
Formaldehyde	831
Di-2-ethylhexyl sebacate	58.5
Di-sec-amyl sebacate	72.9
Di-2-ethylhexyl adipate	67.4

EXPERIMENTAL RESULTS

The Immediate Toxicity of the Materials When Administered Orally

The mortality that resulted when each of the 3 undiluted esters was given in a series of suitably spaced oral dosages to separate groups of either rabbits or rats is shown in Table 1. Comparable data for formulations WS-2211, PRL-3313 and PRL-3039 are presented in Table 2. The ranges within which the minimum lethal doses fell are presented below.

Material	M.L.D. (g/kg of body weight) Rabbits	Rats
Di-2-ethylhexyl adipate	3.2 - 4.7	7.1 - 10.7
Di-2-ethylhexyl sebacate	2.1 - 3.2	10.7 - 16.0
Di-sec-amyl sebacate	4.7 - 7.1	7.1 - 10.7
PRL-3039	16.0 - 24.0	24.0 - 36.0
WS-2211 (unused)(MRD-52-8)	3.2 - 4.7	4.7 - 7.1
WS-2211 (used)(MRD-52-9)	3.2 - 4.7	7.1 - 10.7
PRL-3313	1.4 - 2.1	7.1 - 10.7

No ready explanation can be offered for the difference between the results given by PRL-3039 and that of its principal ingredient, di-sec-amyl sebacate other than the somewhat remote possibility that certain unusually resistant animals were included among those given large dosages of PRL-3039. Perhaps of more importance, as a source of variable results among small numbers of animals, is the fact that these oily materials passed rapidly through the digestive tracts of the animals and so were subject to considerable variability with respect to the extent of their absorption. This may account for the fact that some animals survived after having been given dosages that were lethal for others. Weakness, prostration, ataxia and dyspnea were induced by the administration of these materials.

The oral administration of di-2-ethylhexyl adipate, di-2-ethylhexyl sebacate and di-sec-amyl sebacate in lethal quantities resulted in degeneration of the livers, kidneys and brains of rabbits and rats. Some of the animals given di-2-ethylhexyl sebacate exhibited pulmonary hyperemia and edema. Di-2-ethylhexyl adipate induced necrosis of the mucosal epithelium in the small intestine of rats, and severe gastritis in rabbits.

Sub-lethal doses of di-sec-amyl sebacate induced degenerative changes in the livers and kidneys of rats and in the livers of rabbits, but the viscera of animals given sub-lethal doses of either of the other 2 esters showed no histopathological changes.

The changes induced in the tissues by the 3 formulations were similar, in general, to those elicited by their chief components. PRL-3313, like its chief component, di-2-ethylhexyl sebacate, gave rise to edema and hyperemia of the lungs of some of the animals given a lethal dose, but pulmonary changes were not encountered in animals given either of the other esters or their formulations. Only in the case of animals given a lethal dose of PRL-3313 did the brain show diffuse degenerative lesions.

The Effects of the Oral Administration of Multiple Doses of WS-2211 to Rabbits and Rats

Each of 3 rabbits, given orally the dosage of 0.40 ml of unused WS-2211 (MRD-52-8) per kilogram of body weight on each of 5 days per week, and each of 4

rabbits, given used WS-2211 (MRD-52-9), survived after each had been given 50 doses over the period of 10 weeks (Table 4). Each of 3 rats, given the dosage of 0.90 ml of unused WS-2211 per kilogram of body weight, and each of 4 others, given the used material in the same dosage, survived after having been given 50 doses over the period of 10 weeks. No signs of intoxication were observed. The animals were killed 1 to 15 days after the last dose had been given, at which time microscopic examination of their tissues did not show the presence of any lesions.

The Effects of Contact of the Material With the Skin of Rabbits Over the Period of Twenty-four Hours

The minimum lethal dose of di-2-ethylhexyl adipate, WS-2211 (unused, MRD-52-8), WS-2211 (used, MRD-52-9), and PRL-3039, when maintained in contact with either the intact or abraded skin of rabbits by the 24-hour sleeve method of Draize, et al., is greater than 9.4 ml per kg of body weight (Table 3). When, under corresponding experimental conditions, di-2-ethylhexyl sebacate or di-sec-amyl sebacate remained in contact with the intact skin of rabbits, or PRL-3313 with the abraded skin of rabbits, the minimum lethal dose was also greater than 9.4 ml per kg. However, when either of the sebacates was maintained in contact with the abraded skin of rabbits and PRL-3313 was maintained in contact with the intact skin of rabbits, the minimum lethal dose was greater than 6.0 and less than 9.4 ml per kg. That PRL-3313 appeared to be more toxic when in contact with intact than abraded skin may be regarded as the result of the chance inclusion of one unduly susceptible animal in the small number used for the experiment.

The intact skin of animals subjected to contact with di-2-ethylhexyl adipate or PRL-3039 exhibited a slight erythema, while the abraded skin, in addition to redness, developed a few petechial hemorrhages at the sites of the abrasions. The skin of the animals exposed to contact with the other esters, PRL-3313 or WS-2211 (used or unused), remained unaltered when inspected grossly. In no instance was any sign of systemic intoxication observed.

In general, such transitory losses in weight as occurred among survivors were not much greater than those encountered among control rabbits kept similarly in stocks.

Contact of the various substances with the intact or abraded skin of rabbits induced the following visceral changes:

Di-2-ethylhexyl adipate	- pulmonary hyperemia and edema; no hepatic damage seen
Di-2-ethylhexyl sebacate	- degeneration of the liver
Di-sec-amyl sebacate	- degeneration of the liver, heart, kidneys and brain
PRL-3039	- pulmonary edema and degeneration of the brain and kidneys
PRL-3313	- degeneration of the liver, kidneys and brain, and sub-acute inflammation of the skin
WS-2211	- subacute inflammation of the skin

No great significance should be attached to the differences in the pathological findings associated with the percutaneous absorption of the several materials, since relatively few animals were subjected to any one of them and some of the animals were subjected to lethal and others to non-lethal dosages.

The Effects of Prolonged Intermittent Contact of WS-2211 (MRD-52-8 and MRD-52-9) With the Skin of Rabbits

Table 5 presents the data obtained when undiluted WS-2211 was kept for 2 hours upon the intact skin of rabbits on each of 5 days per week over periods of either 1.5 or 10 weeks. Three rabbits survived following the subjection of each of them to contact with 5.0 ml of unused WS-2211 (MRD-52-8) on each of 8 days over a period of 10 days. Applications upon these animals, which were losing weight, were discontinued at this time because of the condition of the skin, which was thickened and fissured. Similar results were obtained when this dosage of the used material was applied (Table 5). Each of 3 rabbits, given the dose of 2.0 ml of unused WS-2211 on each of 5 days per

week, survived after contact with 50 such doses over the period of 10 weeks. Slight erythema, dryness and slight cracking of the skin were noted more prominently during the earlier, than in the latter, part of the experimental period. The results were the same when the used WS-2211 was applied (Table 5) in like dosage.

Multiple applications of WS-2211 (used or unused) upon the skin of rabbits caused subacute inflammation, hyperkeratosis and acanthosis, but there were no lesions of the viscera. The cutaneous alterations appeared to be of somewhat lesser degree when the lubricant applied had been used previously in an airplane motor.

Exposure to the Vapor of the Esters

Three groups of animals, composed of several species, survived following exposure for 7 hours on each of 5 consecutive days to air containing 0.031 mg of di-2-ethylhexyl adipate per liter, 0.0098 mg of di-2-ethylhexyl sebacate per liter, and 0.025 mg of di-sec-amyl sebacate per liter, respectively (Table 6). No signs of intoxication other than slight transitory losses in weight were observed in these animals. Exposure to these vapors did not result in any demonstrable changes in the gross or microscopic appearance of the organs of the animals.

Exposure to the Undecomposed Mist

Pertinent data concerning the exposure of the animals to the mists of these materials are given in Table 7. A group of animals, consisting of a cat, 2 guinea pigs, 2 rabbits and 4 rats, survived without signs of intoxication, following exposure to the mist of di-2-ethylhexyl sebacate in the air in the concentration of 0.40 mg per liter, for 7 hours on each of 10 days over a period of 12 days. The liver and kidneys of one of the rats were found to have undergone degenerative changes, while lesions of minimal bronchitis and pneumonia were found in the cat. The viscera from all other animals were normal.

Animals of several species survived without signs of intoxication, when exposed to a mist of di-sec-amyl sebacate in the concentration of 0.60 mg per liter, for 7 hours on each of 10 days over a period of 12 days.

There was slight fatty vacuolation of the livers of animals of all the species, except the rabbit. The other viscera were normal.

Except for a rat, a group of animals consisting of 2 guinea pigs, 3 rabbits and 4 rats, survived without untoward reactions, following exposure to the mist of di-2-ethylhexyl adipate in the air in the concentration of 0.93 mg per liter, for 7 hours on each of 10 days over a period of 15 days. Exposure to the mist induced slight degenerative changes of the liver and kidneys of a guinea pig and a rabbit, and slight pneumonitis in a rabbit. Otherwise the viscera of the various species of animals were normal.

Animals of several species survived without signs of intoxication when subjected, for the respective periods of time, to exposure to air bearing a mist of unused WS-2211 containing di-2-ethylhexyl sebacate in the concentrations listed: 70 hours, 1.14 mg per liter; 7 hours, 1.58 mg per liter; and 3.5 hours, 1.43 mg per liter. On microscopic examination, the rabbits and guinea pigs were found to have toxic degenerative changes in their livers, kidneys and adrenal glands and slight pneumonitis. Except for slight pneumonitis the viscera of cats and rats were normal.

Animals of several species survived without signs of intoxication, following exposure to air bearing a mist of used WS-2211 for 7 hours per day on each of 10 days over a period of 12 days. The air contained di-2-ethylhexyl sebacate in the concentration of 0.87 mg per liter. When the tissues of one animal of each species were examined microscopically, the liver and kidneys of the guinea pig and rat were found to have suffered some slight degenerative changes. The organs of the dog and rabbit were normal.

A cat, 2 guinea pigs and 4 rats survived when exposed, for 7 hours on each of 10 days over a total period of 12 days, to air bearing a mist of PRL-3313, but 2 rabbits died of severe pneumonitis after the 9th and 10th periods of exposure, respectively. The air contained di-2-ethylhexyl sebacate in the concentration of 0.72 mg per liter. A similar group survived when subjected, for 7 hours on each of 10 days over a period of 14 days, to air bearing a mist of PRL-3313 in the concentration, expressed in terms of its content of di-2-ethylhexyl sebacate, of 0.16 mg per liter. The cat and the rabbits exhibited salivation, and the

rabbits became prostrate in the first experiment, but no untoward reactions were observed among any of the animals subjected to the lower concentration. Pneumonitis was observed in all species subjected to the mists of this formulation, but only the 2 rabbits that died exhibited other visceral lesions, these consisting of degeneration of the liver and focal degeneration of the renal tubules.

Pneumonitis of varying intensity was observed in animals exposed to the mist of formulations WS-2211 (unused) and PRL-3313, both of which consisted principally of di-2-ethylhexyl sebacate, but no pulmonary lesions were observed in animals which had been exposed to the mist of the principal ingredient alone or to the used formulation of WS-2211. The used WS-2211, when applied from day to day on the skin of rabbits, was shown to be less irritating than the unused WS-2211. Many more animals would have to be employed to demonstrate whether the pneumonitis apparently associated with exposure to the formulations (unused WS-2211 and PRL-3313), while failing to appear under conditions of exposure to their principal ingredient, may properly be regarded as having been induced by the exposure or whether it was of incidental origin. It is not unreasonable to assume, however, that there were differences in the irritant properties of the materials to which the animals were exposed, and certainly it is the better part of prudence to accept the evidence, although it is somewhat dubious, that these substances are potentially capable of inducing pneumonitis, until there is more satisfactory evidence to the contrary.

Exposure to Products Arising from Contact With a Heated Inconel Surface

Di-2-Ethylhexyl Sebacate. The mortality among the several groups of guinea pigs, rabbits and rats exposed to the fog formed by contact of di-2-ethylhexyl sebacate with Inconel at 700° F. has been plotted logarithmically in relation to the severity of exposure in Figures 9, 10 and 11, respectively. In all instances, the rate at which air flowed through the furnace was 31.8 liters per minute. In the graphs, the duration of exposure, in hours, is plotted along the axis of abscissae and the corresponding rate of delivery, in milligrams per minute, along the axis of ordinates. Accordingly, the location of a point anywhere on such a graph indicates the severity of an

exposure, as expressed in terms of the rate of delivery of the ester into the furnace and the length of time the fog in the air was inhaled. The fate of the animals in each experiment has been represented by circles, the extent to which each circle is filled indicating the percentage of animals that died under stated conditions. In Figures 9 and 11, two parallel lines have been drawn; the one to the left separates non-lethal conditions from those associated with the death of some but not all of the group, while the one to the right separates the latter from uniformly lethal conditions of exposure. In Figure 10, a single line has been drawn to separate the non-lethal conditions from those associated with the death of some but not all of the group.

Rabbits and rats (Figures 10 and 11) exposed to the fog for 24 hours survived longer than might have been anticipated had the original lines been extended without the change in slope made necessary by the data. Such a deflection indicates that the animals were exposed to a concentration of such character that the duration of exposure was relatively unimportant in relation to the fate of the animals.

It is readily apparent that guinea pigs are more resistant than rabbits or rats (Table 8). Five of 24 (20.8 per cent) guinea pigs, 17 of 47 (36.2 per cent) rabbits, and 20 of 48 (41.7 per cent) rats died when exposed to fogs formed from di-2-ethylhexyl sebacate at 700°F.

All guinea pigs and rabbits and all but 1 of 4 rats survived when exposed for 2 hours to the fog formed by dropping 56 mg. of di-2-ethylhexyl sebacate per minute into an Inconel tube, heated to 700°F, through which air was passing at the rate of 31.8 liters per minute (Table 8). As the rates of delivery increased from 85 to 223 mg per minute, while other factors were held constant, the incidence of mortality in other like groups increased abruptly. Animals that died had acute chemical pneumonitis and degeneration of the brain, liver and kidneys. Those that survived had normal viscera.

When the liquid was added to the Inconel at the rate of 56 mg per minute, the air of the chamber contained total aldehydes, expressed as formaldehyde, in the concentration of 60 p.p.m., formaldehyde, alone, in that of 8.2 p.p.m., and ester, expressed as di-2-ethylhexyl sebacate, in the concentration of 0.62 mg

per liter. When the ester was delivered at the rate of 85 mg per minute, some of the animals died and the concentrations of these substances in the chamber were, respectively, 139 p.p.m., 32 p.p.m., and 1.22 mg per liter. Carbon monoxide was present also, to the extent of 348 p.p.m.

All of the animals survived exposure for 7 hours to the fog formed by dropping the ester at the rate of either 14.1 or 19.6 mg per minute into an Inconel tube heated to 700°F (Table 8). Increases in the rate of delivery of the liquid upon the heated metal to 30.1, 37 or 61 mg per minute caused sharp increases in the incidence of mortality among rabbits and rats, but in only one instance (61 mg per minute) did a guinea pig die. Animals that died had acute chemical pneumonitis and degeneration of the brain, liver and kidneys. Animals that survived had only residual chemical pneumonitis.

The highest concentrations of the respective products of decomposition which permitted survival of all the animals, following exposure for 7 hours, were: total aldehydes, 32 p.p.m.; formaldehyde, 5.3 p.p.m.; carbon monoxide, 51 p.p.m.; and ester, 0.23 mg per liter. The lowest concentrations in the chamber that were lethal to half the group were, respectively: total aldehydes, 42 p.p.m.; formaldehyde, 7 p.p.m.; carbon monoxide, 63 p.p.m.; and ester, 0.36 mg per liter.

When animals were subjected to exposure for 24 hours to the thermal decomposition products arising when di-2-ethylhexyl sebacate was delivered at the rate of either 7.4 or 16.5 mg per minute into the Inconel tube, heated to the temperature of 700°F (Table 8), all, except one rat exposed to the higher level, survived. Inhalation of the products under the conditions described did not induce any significant visceral lesions.

When the experiments continued over 24-hour periods, the concentrations of the respective contaminants were found to be: total aldehydes, 12 and 26 p.p.m., formaldehyde, 2.7 and 4.6 p.p.m., carbon monoxide, 30 and 39 p.p.m., and ester, 0.07 and 0.12 mg per liter.

The fogs of di-2-ethylhexyl sebacate produced irritation of the mucous membranes and dyspnea.

There appears to be a critical temperature between 550°F (288°C) and 600°F (316°C) above which di-2-ethylhexyl sebacate is decomposed to form a toxic fog. A limited number of experiments at 600°F (Table 10) and at 900°F (Table 9) reveal no difference in the toxicity of the fogs formed at either temperature from that of a fog formed at 700°F; however, the fogs formed at 550° and 400°F (Table 10) were much less toxic than those formed at 700°F.

Inhalation of the fogs formed at 600° and at 900°F induced an acute chemical pneumonitis and diffuse degeneration of the brain, liver and kidneys in all animals that died.

Inhalation of the fogs formed at 550° and 400°F did not induce any significant alterations of the viscera of any of the animals, with the one exception of a rabbit that died in the experiment which involved the higher rate of delivery. This animal had acute pneumonitis and degeneration of the liver and kidneys.

The amount of ester found in the air of the chamber was related to the amount dropped upon the Inconel at 700°F. With increase in the temperature of the Inconel tube into which the ester was dropped, the extent of the total recovery of the ester diminished, but the proportion of the ester which was recovered in the form of vapor increased.

Di-sec-Amyl Sebacate. The mortality in the several groups of guinea pigs, rabbits and rats exposed to the fog formed by contact of di-sec-amyl sebacate with Inconel at 700°F has been plotted logarithmically in relation to the severity of exposure in Figures 12, 13 and 14, respectively.

It is apparent that guinea pigs were more resistant than rabbits or rats (Table 11) to di-sec-amyl sebacate. One of 20 (5.0 per cent) guinea pigs, 14 of 40 (35.0 per cent) rabbits and 16 of 40 (40.0 per cent) rats died when exposed to the fogs formed from di-sec-amyl sebacate.

The threshold rate of delivery of di-sec-amyl sebacate into the Inconel tube heated to 700°F for the formation of a lethal fog, under the specified experimental conditions (2 hours of exposure with air flowing through the Inconel tube at the rate of 31.8 liters per minute), was between 92 and 127 mg per minute in the case of rabbits and rats, and above 252 mg per minute

in the case of guinea pigs (Table 11). Inhalation of the fog by rabbits and rats induced a severe chemical pneumonitis and degeneration of the liver, kidney and brain. Guinea pigs exhibited only slight degenerative changes in the liver. The air of the chamber contained the following contaminants in the stated concentrations, when the rates of delivery were 92 and 127 mg per minute, respectively: total aldehydes, expressed as formaldehyde, 61 and 98 p.p.m.; formaldehyde, alone, 9 and 10 p.p.m.; carbon monoxide, 39 and 46 p.p.m.; and ester, expressed as di-sec-amyl sebacate, 0.76 and 0.93 mg per liter.

All animals, except a rabbit and a rat, survived when exposed for 7 hours to fogs formed by dropping either 19.0 or 32.2 mg of the sebacate per minute into an Inconel tube heated to 700°F. Increase in the rate of delivery of the liquid upon the heated Inconel (700°F) to 44.3 or to 65 mg per minute caused sharp increase in the incidence of mortality among rabbits and rats. In only one of the series of 10 experiments did a guinea pig die (Table 11). Exposure for 7 hours to the fogs formed at the higher rates of delivery (44.3 or 65 mg per minute) induced slight degenerative changes in the liver and kidneys of all species of animals, but no significant visceral lesions were found in any animal when the fog was formed at the lower rates of delivery (19 or 32 mg per minute). The concentrations associated with rates of delivery of 32.2 and 44.3 mg per minute, respectively, were: total aldehydes, 33 and 51 p.p.m., formaldehyde, 10 and 7.5 p.p.m., carbon monoxide, 29 and 31 p.p.m., and ester, 0.38 and 0.39 mg per liter.

When animals were subjected to exposure for 24 hours to the thermal decomposition products arising when di-sec-amyl sebacate was delivered at the rate of 19.5 mg per minute into the Inconel tube heated to 700°F, 2 of 4 rats died, and 4 rabbits and 2 guinea pigs survived (Table 11). However, all of the exposed animals survived when the liquid was added to the Inconel at the rate of 8.4 mg per minute. The viscera of most of these animals were normal, although slight pneumonitis and slight degeneration of the liver were noted in one animal of each species exposed to the higher of these two concentrations.

When experimental conditions were such that exposure over the period of 24 hours was lethal, the concentrations of the respective contaminants were

found to be: total aldehydes, 23 p.p.m., formaldehyde, 3.0 p.p.m., carbon monoxide, 21 p.p.m., and ester, 0.26 mg per liter. When the conditions were such that exposure over this period could be tolerated, the concentrations were, respectively: total aldehydes, 7.5 p.p.m., formaldehyde, 0.2 p.p.m., carbon monoxide, 12 p.p.m., and ester, 0.07 mg per liter.

The critical temperature for the generation of a toxic fog appeared to lie between 550° and 600°F, since fogs obtained at either 600° (Table 13) and 900°F (Table 12) gave results comparable to those obtained at 700°F, while that formed at 550°F (Table 13) did not induce fatalities. Exposure to the fogs at 600° or 900°F induced degeneration of the brain, liver and kidney, and pulmonary irritation in rabbits and rats. Exposure to the fogs at 550°F induced no lesions of the viscera.

Lethal conditions were associated with weakness and respiratory distress among the animals, whereas only escape reactions and irritation of the mucous membranes were observed under non-lethal conditions.

Although the incidence of mortality among guinea pigs, rabbits and rats was slightly greater when the fog of di-2-ethylhexyl sebacate was generated at 700°F than when di-sec-amyl sebacate was added to the Inconel under comparable conditions, the differences were not statistically significant.

Di-2-Ethylhexyl Adipate. The mortality in the several groups of cats, guinea pigs, rabbits and rats exposed to the fog formed by contact of di-2-ethylhexyl adipate with Inconel at 700°F have been plotted logarithmically in relation to the severity of exposure in Figures 15, 16, 17 and 18, respectively.

As in the case of the sebacate esters the guinea pigs were the least susceptible of the species employed (Table 14). Six of 30 (20.0 per cent) guinea pigs, 4 of 13 (30.8 per cent) cats, 18 of 32 (56.3 per cent) rabbits, and 29 of 60 (48.3 per cent) rats died when exposed to the fogs formed from di-2-ethylhexyl adipate at 700°F.

The highest tolerable dosage and the lowest lethal dosage, expressed in terms of milligrams of di-2-ethylhexyl adipate delivered into an Inconel tube maintained at 700°F through which air was passing at the rate of

31.8 liters per minute, are given below for the several species of animals and the stated duration of exposure:

Species of Animal	mg per minute Tolerated	Lethal
	2 hours	
Cats	173.5	218.0
Guinea pigs	116.7	173.5
Rabbits	49.2	116.7
Rats	116.7	173.5
	7 hours	
Cats	49.5	54.0
Guinea pigs	54.0	64.1
Rabbits	24.8	49.5
Rats	24.8	49.5
	24 hours	
Cats	10.7	-
Guinea pigs	10.7	-
Rabbits	7.2	10.7
Rats	7.2	10.7

Exposure to the fog formed at 700°F induced acute diffuse chemical pneumonitis, and degenerative changes of the brain, liver and kidneys in all species of animals exposed. Animals that survived had similar visceral lesions of a lesser degree of severity.

The fog formed from the adipate in contact with the Inconel at 500°F (Table 15) was of the same order of toxicity as that formed at 700°F. However, only 1 of 36 animals died when exposed to the fog arising from the Inconel at 400°F (Table 16). The tissue changes induced by exposure of animals to the fog formed at 500°F were similar to those caused by exposure to the fog formed at 700°F. However, exposure to the fog formed at 400°F did not induce any significant visceral lesions. The death of one of the guinea pigs which were subjected to the conditions of the latter experiment (at 700°F) resulted from an infectious pneumonia which is believed to have been incidental.

The denser fogs formed at the higher temperatures (500° and 700°F) induced moderate to severe irritation

of the mucous membranes and dyspnea, but those formed at 400°F induced only slight irritation of the mucous membranes.

Small quantities of aldehydes and carbon monoxide were present, but only occasionally was formaldehyde detected among the aldehydes present. Large quantities of finely divided droplets of liquid were present.

The toxicity of the fogs of di-2-ethylhexyl adipate formed at 700° or 400°F was comparable in degree to that of the fog formed from di-2-ethylhexyl sebacate at the corresponding temperatures. However, the critical temperature for the production of a toxic fog appeared to be lower in the case of the adipate than in that of the sebacate, the fog from the former at 500°F (Table 15) seeming to be more toxic than that formed from the latter at 550°F (Table 10).

Formulation WS-2211. The mortality in the several groups of guinea pigs, rabbits and rats exposed to the fog formed by contact of WS-2211 (unused) with Inconel at 700°F has been plotted logarithmically in relation to the severity of exposure, in Figures 19, 20 and 21, respectively.

As in the case of the 3 esters referred to previously, the guinea pigs were the most resistant of the animals employed in these experiments (Table 17). One of 24 (4.2 per cent) guinea pigs, 1 of 3 (33.3 per cent) cats, 19 of 32 (59.4 per cent) rabbits and 19 of 42 (45.2 per cent) rats died following exposure to the fogs arising from contact of WS-2211 with Inconel maintained at 700°F.

All animals, except one rabbit, survived when exposed for 2 hours to fogs formed by dropping either 45.0 or 60.0 mg of WS-2211 per minute into an Inconel tube, heated to 700°F, through which air was passing at the rate of 31.8 liters per minute (Table 17). As the rates of delivery of the liquid were increased to 86.7 mg per minute and higher, while other factors were held constant, the incidence of mortality among rabbits and rats increased abruptly, but guinea pigs survived without exception even when the liquid was delivered at the rate of 207 mg per minute. The animals of various species exposed for 2 hours to fogs formed by dropping WS-2211 into the heated Inconel tube were found to have microscopic degenerative changes in their viscera as follows: rabbits - degeneration of brain,

liver and kidneys; rats - degeneration of brain, liver, kidneys, heart and adrenal glands; guinea pigs - focal pneumonia, degeneration of liver and adrenal glands; cat - degeneration of the brain, liver and kidneys. Pulmonary irritation in rabbits, rats and a cat was characterized by acute bronchitis and pulmonary edema and hyperemia.

When the liquid was delivered at the rate of 45.0 mg per minute, the air of the chamber contained aldehydes, expressed as formaldehyde, in the concentration of 56.5 p.p.m., formaldehyde, alone, in that of 7.6 p.p.m., and carbon monoxide in that of 21.4 p.p.m. The lethal concentrations of these substances found when the delivery rate was 86.7 mg of WS-2211 per minute were, respectively, 70.6 p.p.m., 19.2 p.p.m. and 329.6 p.p.m.

All animals, except one rabbit, survived when exposed for 7 hours to fogs formed by dropping either 15.0 or 19.5 mg of WS-2211 per minute into an Inconel tube. Increase in the rate of delivery of the liquid upon the heated metal to 30, 43 or 61 mg per minute caused a sharp increase in the incidence of mortality among rabbits and rats. In only one instance (61 mg per minute) did a guinea pig die (Table 17). The animals exposed for 7 hours to fogs formed from WS-2211 exhibited degeneration of the brain, liver and kidneys, and, in some instances, of the heart and adrenal glands. Pulmonary irritation in all species was manifested by acute bronchitis and pulmonary edema and hyperemia.

The highest concentrations of the respective products of decomposition that were associated with the survival of all but one of the animals, following exposure for 7 hours, were: total aldehydes, 19.9 p.p.m.; formaldehyde, 3.1 p.p.m.; and carbon monoxide, 66.8 p.p.m. The lowest concentrations in the chamber in experiments that resulted in the death of more than half the group were, respectively: total aldehydes, 39.8 p.p.m.; formaldehyde, 6.3 p.p.m.; and carbon monoxide, 114 p.p.m.

When animals were subjected to exposure for 24 hours to the thermal decomposition products arising when WS-2211 was delivered at the rate of 17.9 mg per minute into the heated Inconel tube, the incidence of mortality among rabbits and rats was high, but all of the guinea pigs survived. When the rate was decreased to 7.4 mg of WS-2211 per minute, all of the rabbits and

rats, as well as the guinea pigs, survived (Table 17). Exposure for 24 hours to fogs generated from WS-2211 resulted in degeneration of the liver and kidneys of all species. Guinea pigs, rats and rabbits showed only slight pneumonitis.

When the experimental conditions over 24-hour periods of exposure were lethal, the concentrations of the respective contaminants were found to be: total aldehydes, 31.9 p.p.m.; formaldehyde, 4.5 p.p.m.; and carbon monoxide, 60.6 p.p.m. When the conditions could be tolerated over this same period, the concentrations were, respectively: total aldehydes, 13.0 p.p.m.; formaldehyde, 1.7 p.p.m.; and carbon monoxide, 40.1 p.p.m.

The fog of decomposition products induced considerable irritation of the mucous membranes, even under the least severe experimental conditions observed.

Under comparable conditions of exposure, the fog formed from WS-2211 at 400°F caused fewer animals to die than did the fog formed at 700°F. All of the animals survived, except one rabbit and one rat, when 5 groups, each consisting of 2 guinea pigs, 4 rats and either 3 rabbits or 2 rabbits and a cat, were exposed for 7 hours to the fog formed by dropping WS-2211 at rates within the range of 23.0 to 83.6 mg per minute, into an Inconel tube heated to 400°F, through which air was passing at the rate of 31.8 liters per minute (Table 18). Aside from some salivation on the part of occasional animals, no signs of intoxication were observed.

All animals survived, except a rabbit and a rat, when 2 guinea pigs, 3 rabbits and 4 rats were exposed for 7 hours to the fog formed by dropping 32.9 mg of WS-2211 per minute into an Inconel tube, maintained at the temperature of 550°F (Table 18). The fog formed at 550°F, which induced irritation of the exposed mucous membranes of the animals, was less toxic than that formed at 700°F, and apparently only slightly more toxic than that formed at 400°F, although the atmosphere contained total aldehydes, formaldehyde and carbon monoxide in concentrations as great as those which resulted from the higher temperature of 700°F.

The atmospheric concentrations of the decomposition products formed at 400°F varied with the rates of delivery of the oil into the tube, but the concentrations of total aldehydes and of formaldehyde, which, in

the several experiments, ranged from 0.3 to 24 p.p.m., and from 0.0 to 2.1 p.p.m., respectively, were much lower than those formed at the higher temperature of 700°F. Under the conditions represented by the temperature of 400°F, the concentration of carbon monoxide ranged from 63 to 481 p.p.m. and that of esters, expressed as di-2-ethylhexyl sebacate, from 0.14 to 0.74 mg per liter.

Formulation PRL-3039. The mortality in the several groups of guinea pigs, rabbits and rats which were exposed to the fog formed by contact of PRL-3039 with Inconel at 700°F, has been plotted logarithmically in relation to the severity of exposure in Figures 22, 23 and 24, respectively.

Mortality, which was a function of the duration of exposure and the rate of delivery of PRL-3039 into the Inconel, varied from one species to another, the susceptibility decreasing in the following order: rabbits, rats and guinea pigs. When decomposition occurred at 700°F, 19 of 40 (47.5 per cent) rabbits, 8 of 40 (20.0 per cent) rats, and 1 of 19 (5.3 per cent) guinea pigs died (Table 19). Deaths occurred among animals of various species exposed for the stated period at the higher rates of delivery of the liquid into the furnace (700°F) and animals survived uniformly at the lower rates.

Species of Animal	mg of PRL-3039 per minute		
	Two Hours	Seven Hours	Twenty-four Hours
Guinea pigs	62- 91(1)	65 ->65	19.3->19.3
Rabbits	62- 91	19.4- 30.1	<7.3- 7.3
Rats	91-114	45 - 65	19.3->19.3

(1) The death of a guinea pig was unexpected.

The fogs formed from PRL-3039 at 900°F (Table 20) were much more toxic for rats than those formed at 700°F, but were only slightly more toxic for rabbits, and not demonstrably more toxic for guinea pigs, all of which survived. Respiratory distress and delayed deaths were common among the animals employed in this experiment. Animals that died exhibited acute chemical pneumonitis and degenerative changes of the liver and kidneys. Animals that survived following exposure to the more severe conditions of exposure had residual pneumonitis and degeneration of the liver and kidneys.

When the formulation was decomposed at the lower temperature (700°F) the air in the chamber contained total aldehydes, expressed as formaldehyde, in the concentrations of 22 - 376 p.p.m., formaldehyde in the concentration of 3 to 56 p.p.m., carbon monoxide in the concentrations of 18 to 65 p.p.m., and carbon dioxide in the concentrations of 2,948 to 5,103 p.p.m. Total particulate material, which was largely undecomposed sebacate ester with some free acid, was present from 0.10 to 1.45 mg per liter. The concentrations of total particulate matter, total aldehydes and formaldehyde were related to the rate of delivery of PRL-3039 into the furnace, but this relationship was somewhat variable in the case of the oxides of carbon. Except in the case of carbon monoxide, the concentrations of these contaminants were slightly lower in comparable fogs formed at 900°F. At 900°F a greater percentage of the particulate matter was present as free acid.

DISCUSSION

According to the terminology employed by Hodge and Sterner, these esters and their formulations may be classified as only slightly toxic or practically non-toxic, when given orally to animals. Likewise, they may be regarded as practically non-toxic when in contact with the skin of rabbits.

The hazards associated with the undecomposed mists of the esters or of formulations WS-2211 or PRL-3313 should be small, since concentrations such as those to which animals were exposed in these experiments would be encountered but rarely.

The thermal decomposition of these materials increases their toxicity considerably. There appears to be a critical temperature above which toxic products begin to form. In the case of di-2-ethylhexyl sebacate, di-sec-amyl sebacate, and formulation WS-2211 this critical temperature was between 550° and 700°F, whereas in the case of di-2-ethylhexyl adipate it was between 400° and 500°F, and in that of formulation PRL-3039 it was less than 700°F.

The toxicity of the fogs formed from these esters and their formulations, when dropped into an Inconel tube maintained at 700°F, are of the same order of magnitude. The toxicity of formulation WS-2211 is largely

that of its principal ingredient, di-2-ethylhexyl sebacate, while the tricresyl phosphate contributes little or nothing to its toxicity. In investigations sponsored by Monsanto Chemical Company, of the thermal decomposition of a commercial grade of mixed tricresyl phosphates, containing a low content of the ortho isomer, guinea pigs were the most susceptible animals employed, whereas in all of these experiments with adipates or sebacates, guinea pigs have been the most resistant. The work carried out on tricresyl phosphate has been reported in WADC Technical Report No. 54-345.

The toxicity of the thermal decomposition products of these aliphatic esters and their formulations is not greatly different from that of the thermal decomposition products of a typical paraffinic hydrocarbon fluid. For purposes of comparison, the results of an investigation of the paraffinic hydrocarbon, carried out under the sponsorship of Monsanto Chemical Company, have been plotted with those reported herein for di-2-ethylhexyl sebacate. The mortality in the several groups of guinea pigs, rabbits and rats are shown in Tables 25, 26 and 27, respectively.

SUMMARY

Di-2-ethylhexyl sebacate, di-sec-amyl sebacate, di-2-ethylhexyl adipate, and formulations WS-2211 and PRL-3313, when given undiluted in a single oral dose to animals are either slightly toxic or practically non-toxic. The esters and formulations WS-2211 and PRL-3039 when maintained in contact with either the intact or abraded skin of rabbits for 24 hours are practically non-toxic. Repeated applications of large doses of WS-2211 upon the skin of rabbits resulted in irritation.

High concentrations of the mists of these substances were generally tolerated without untoward reactions.

When animals of several species were exposed for stated periods to air bearing fogs formed by the thermal decomposition of these materials at 700°F, the highest tolerable dosage and the lowest lethal dosage, expressed in milligrams of the substance delivered into

a heated furnace per liter of air passing through it, were as given below for the species found most susceptible.

Material	Species	Atmospheric Concentration mg/liter: Tolerated	Atmospheric Concentration mg/liter: Lethal to Some of the Group
Duration of Exposure: 2 hours			
Di-2-ethylhexyl sebacate	rats	-	1.76
Di-sec-amyl sebacate	rabbits rats	2.89	3.99
Di-2-ethylhexyl adipate	rabbits	1.55	3.67
WS-2211	rabbits	-	1.42
PRL-3039	rabbits	1.95	2.86
Duration of Exposure: 7 hours			
Di-2-ethylhexyl sebacate	rabbits rats	0.62	0.95
Di-sec-amyl sebacate	rabbits	-	0.60
Di-2-ethylhexyl adipate	rabbits rats	0.78	1.56
WS-2211	rabbits	-	0.47
PRL-3039	rabbits	0.61	0.95
Duration of Exposure: 24 hours			
Di-2-ethylhexyl sebacate	rats	0.23	0.52
Di-sec-amyl sebacate	rats	0.26	0.61
Di-2-ethylhexyl adipate	rabbits rats	0.23	0.34

Material	Species	Atmospheric Concentration mg/liter	
		Tolerated	Lethal to Some of the Group
WS-2211	rabbits rats	0.23	0.56
PRL-3039	rabbits	-	0.23

BIBLIOGRAPHY

Bland, D. E. A Micromethod for the Determination of Carbon Monoxide in the Blood. Australian J. Exptl. Biol. Med. Sci., Vol. 18, 1940. pp. 35-47.

Bricker, C. E. and Johnson, H. R. Spectrophotometric Method for Determining Formaldehyde. Ind. Eng. Chem. Anal. Ed., Vol. 17, June, 1945. pp. 400-402.

Draize, J. H., Woodard, G. and Calvery, H. O. Methods for the Study of Irritation and Toxicity of Substances Applied Topically to the Skin and Mucous Membranes. J. Pharmacol. Exper. Therap., Vol. 82, December, 1944. pp. 377-390.

Goldman, F. H. and Yagoda, H. Collection and Estimation of Traces of Formaldehyde in Air. Ind. Eng. Chem. Anal. Ed., Vol. 15, June, 1943. pp. 377-378.

Hodge, H. D. and Sterner, J. H. Tabulation of Toxicity Classes. Ind. Hyg. Assoc. Quart., Vol. 10, December, 1949. pp. 4-7.

Lundegårdh, H. and Philipson, T. The Spark-in-Flame Method for Spectral Analysis. Lantbruks-Högskol. Ann., Vol. 5, 1938. pp. 249-260. (Swedish) (Chem. Abst. Vol. 32, 1938. p. 6576).

Treon, J. F., Dutra, F. R., Cappel, J., Sigmon, H. and Younker, W. Toxicity of Sulfuric Acid Mist. Arch. Ind. Hyg. Occup. Med., Vol. 2, December, 1950. pp. 716-734.

Willard, H. H. and Furman, N. H. Elementary Quantitative Analysis. Third Edition, D. VanNostrand Co., Inc., New York, 1940. p. 152.

APPENDIX

The methods used and results obtained on the materials described in this report were given in greater detail in the following interim reports:

November 10, 1952 - The Immediate Toxicity of Certain Aliphatic Esters

May 11, 1953 - The Immediate Toxicity of an Aviation Gasoline and Several Hydraulic Fluids That Contain Tricresyl Phosphate

June 16, 1953 - The Toxicity of the Mists Generated by Aspiration from Unused and Used WS-2211 and of the Fog Formed by Dropping Unused WS-2211 Into an Inconel Furnace Heated to Seven Hundred Degrees Fahrenheit

October 30, 1953 - The Toxicity of the Fogs Formed by Dropping WS-2211 Into an Inconel Tube Heated to a Temperature of Four Hundred or Five Hundred Degrees Fahrenheit

March 30, 1954 - The Toxicity of the Mist Generated by Aspiration from Di-2-Ethylhexyl Sebacate and of the Fog Formed by Dropping Di-2-Ethylhexyl Sebacate Into an Heated Inconel Tube

April 10, 1954 - The Toxicity of the Mist Generated by Aspiration from Di-sec-Amyl Sebacate and of the Fog Formed by Dropping It Into an Heated Inconel Tube

April 23, 1954 - The Toxicity of the Mists Generated by Aspiration of PRL-3313

April 27, 1954 - The Toxicity of the Mist Generated by Aspiration from Di-2-Ethylhexyl Adipate and of the Fog Formed by Dropping It Into a Heated Inconel Tube

April 28, 1954 - The Toxicity of the Fog Formed by Dropping PRL-3039 Into a Heated Inconel Tube

April 29, 1954 - The Immediate Toxicity of Synthetic Lubricants MRD-52-8 and MRD-52-9 (Prepared for The Standard Oil Development Company by the Kettering Laboratory)

Table 1

The Immediate Toxicity of Certain Aliphatic Esters When Given Orally to Rabbits and Rats

Dosage ml/kg	Number of Animals that Died/Number Given the Material					
	Di-2-Ethylhexyl Adipate		Di-2-Ethylhexyl Sebacate		Di-sec-Amyl Sebacate	
	Rabbits	Rats	Rabbits	Rats	Rabbits	Rats
36.0	-	1/1	-	2/4	3/3	2/4
24.0	-	4/4	-	4/4	0/3	0/4
16.0	1/1	2/4	-	2/4	2/3	1/4
10.7	1/1	1/4	1/1	0/4	1/3	1/4
7.1	1/1	0/4	3/3	-	1/3	0/4
4.7	2/3	-	1/3	-	0/3	-
3.2	0/3	-	1/3	-	-	-
2.1	0/3	-	0/3	-	-	-

Table 2

The Immediate Toxicity of Formulations Containing Certain Aliphatic Esters When Given Orally to Rabbits and Rats

Dosage ml/kg	Number of Animals that Died/Number Given the Material							
	WS-2211 (Unused) (MRD-52-8)		WS-2211 (Used) (MRD-52-9)		PRL-3313		PRL-3039	
	Rabbits	Rats	Rabbits	Rats	Rabbits	Rats	Rabbits	Rats
36.0	-	-	-	-	-	1/1	0/3	1/4
24.0	-	-	-	-	-	1/1	1/3	0/4
16.0	-	2/4	-	2/4	-	3/4	0/3	0/4
10.7	3/3	1/4	-	1/4	-	1/4	0/3	0/5
7.1	1/3	1/4	3/3	0/4	1/1	0/4	0/3	-
4.7	2/3	0/4	1/3	0/4	3/3	0/4	-	-
3.2	0/3	0/4	0/3	0/4	2/3	0/4	-	-
2.1	0/1	0/4	0/1	0/4	1/3	-	-	-
1.4	-	-	-	-	0/3	-	-	-

Table 3

The Immediate Toxicity of Certain Aliphatic Esters and Certain Formulations Containing Them When Maintained for Twenty-four Hours in Contact With the Skin of Rabbits

Material	Condition of Skin	Number of Animals that Died/Number Given the Material	
		9.4 ml/kg	6.0 ml/kg
Di-2-Ethylhexyl Adipate	Intact Abraded	0/3 0/3	- -
Di-2-Ethylhexyl Sebacate	Intact Abraded	0/3 1/2	- 0/3
Di-sec-Amyl Sebacate	Intact Abraded	0/3 2/3	- 0/3
WS-2211 (Unused) (MRD-52-8)	Intact Abraded	0/3 0/3	- -
WS-2211 (Used) (MRD-52-9)	Intact Abraded	0/3 0/3	- -
PRL-3313	Intact Abraded	1/3 0/3	0/3 -
PRL-3039	Intact Abraded	0/3 0/3	- -

Table 4

The Effects of the Administration of Multiple Oral Doses of a Solution of Either Unused WS-2211 (MRD-52-8) or Used WS-2211 (MRD-52-9) in Peanut Oil to Female Rabbits and Rats

Material	Species of Animal	Concentration of Material in Peanut Oil Per Cent (V/V)	Daily Dose ml/kg		Number of Doses	Average Initial Weight kg	Average Changes in Weight Expressed as Percentage of Initial Weight	Incidence of Mortality
			WS-2211	Solution				
WS-2211 (unused)	Rabbits	10.0	0.40	4.0	50	2.577	+28.0	0/3
WS-2211 (unused)	Rats	25.0	0.90	3.6	50	0.183	+30.6	0/3
WS-2211 (used)	Rabbits	10.0	0.40	4.0	50	2.443	+36.6	0/4
WS-2211 (used)	Rats	25.0	0.90	3.6	50	0.193	+29.3	0/4

Table 5

The Fate of Rabbits When Used (MRD-52-9) or Unused (MRD-52-8) WS-2211 Was Applied Repetitively Without a Diluent Upon Their Skin

Material Applied	Daily Dosage			Number of Doses Per Animal	Weight Change During Period of Exposure, Expressed as Percentage of Initial Weight		Incidence of Mortality
		ml/kg					
	ml	Mean	Range		Mean	Range	
WS-2211 (Unused)	5.0	1.91	1.74-2.02	8	- 5.6	- 0.1 to -11.7	0/3
WS-2211 (Unused)	2.0	0.70	0.53-0.98	50	+39.3	+37.6 to +41.3	0/3
WS-2211 (Used)	5.0	1.93	1.76-2.14	8	- 4.7	- 1.4 to - 9.4	0/3
WS-2211 (Used)	2.0	0.71	0.52-0.95	50	+25.9	+22.0 to +30.8	0/3
None	0.0	-	-	50	+22.8	+14.9 to +33.5	0/3

Table 6

The Fate of Animals Following Exposure to the Vapor of Certain Aliphatic Esters in Air

Material	Concentration		Duration of Exposure Hours	Number of Animals that Died/Number of Animals Exposed				
	mg/l	p.p.m.		Cats	Guinea Pigs	Mice	Rabbits	Rats
Di-2-Ethylhexyl Adipate	0.031	2.1	5 x 7.0	-	0/2	0/5	0/3	0/3
Di-2-Ethylhexyl Sebacate	0.0098	0.57	5 x 7.0	-	0/2	0/5	0/3	0/3
Di-sec-Amyl Sebacate	0.025	1.8	5 x 7.0	0/1	0/2	0/5	0/2	0/3

Table 7

The Fate of Animals Following Exposure to Mists of Certain Aliphatic Esters or Formulations of the Esters in the Air

Material	Amount of Material Aspirated mg/min	Gauge Pressure p.s.i.	Rate of Passage of Air Through Aspirator (at Room Conditions) l/min	Supplementary Flow of Air Into Chamber l/min
Di-2-Ethylhexyl Sebacate	38.2	4	17.6	0
Di-sec-Amyl Sebacate	130.8	4	18.6	41.8
Di-2-Ethylhexyl Adipate	87.5	37	47	0
WS-2211 (Unused)	151.0	38	44 - 48	0
WS-2211 (Unused)	155.5	38	43 - 48	0
WS-2211 (Unused)	94.5	46	46	0
WS-2211 (Used)	81.1	38	47 - 48	0
PRL-3313	77.5	22	11.5	0
PRL-3313	33.8	5	5.6	31.2

(1) Expressed as di-2-ethylhexyl sebacate.

Table 7

The Fate of Animals Following Exposure to Mists of Certain Aliphatic Esters or Formulations of the Esters in the Air

Concentration Found in Chamber mg/l	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed				
		Dogs	Cats	Guinea Pigs	Rabbits	Rats
0.40	10 x 7.0	-	0/1	0/2	0/2	0/4
0.60	10 x 7.0	-	0/1	0/2	0/2	0/4
0.93	10 x 7.0	-	-	0/2	0/3	1/4
1.43 (1)	3.5	-	0/1	0/2	0/2	0/2
1.58 (1)	7.0	-	0/1	0/2	0/2	0/2
1.14 (1)	10 x 7.0	-	0/1	0/2	0/2	0/2
0.87 (1)	10 x 7.0	0/1	-	0/2	0/2	0/2
0.72 (1)	10 x 7.0	-	0/1	0/2	2/2	0/4
0.16 (1)	10 x 7.0	-	0/1	0/2	0/2	0/4

Table 8

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Di-2-Ethylhexyl Sebacate Into an Inconel Tube Heated to the Temperature 700°F (371°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Animals that Died /Number of Animals Exposed		
		Guinea Pigs	Rabbits	Rats
223 (1)	2.0	2/2	3/3	4/4
121	2.0	1/2	4/4	2/4
85	2.0	1/2	3/4	0/4
56	2.0	0/2	0/4	1/4
61	7.0	1/2	3/4	4/4
37	7.0	0/2	2/4	4/4
30.1	7.0	0/2	2/4	3/4
19.8	9.5	0/2	0/4	1/4
19.6	7.0	0/2	0/4	0/4
14.1	7.0	0/2	0/4	0/4
16.5	24.0	0/2	0/4	1/4
7.4	24.0	0/2	0/4	0/4
Total		5/24	17/47	20/48
Percentage		20.8	36.2	41.7

(1) A cat, which was exposed in this experiment, died.

Table 9

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Di-2-Ethylhexyl Sebacate Into an Inconel Tube Heated to 900°F (482°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Animals that Died /Number of Animals Exposed		
		Guinea Pigs	Rabbits	Rats
82	2.0	1/2	3/4	2/4
57	2.0	0/2	0/4	0/4
24	7.0	0/2	0/4	0/4
Total		1/6	3/12	2/12
Percentage		16.7	25.0	16.7

Table 10

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Di-2-Ethylhexyl Sebacate Into an Inconel Tube Heated to 400°F (204°C), 550°F (288°C) or 600°F (316°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Temperature of Inconel °F	Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Animals that Died /Number of Animals Exposed		
			Guinea Pigs	Rabbits	Rats
600	58	7.0	0/2	4/4	3/4
550	57	7.0	0/2	1/4	0/4
550	42.2	7.25	0/2	0/4	0/4
400	36.4	7.0	0/2	0/4	0/4
Total			0/8	5/16	3/16
Percentage			0.0	31.3	18.9

Table 11

The Fate of Animals Following Exposure to Fogs Formed by Dropping Di-sec-Amyl Sebacate Into an Inconel Tube Heated to the Temperature of 700°F (371°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Animals that Died /Number of Animals Exposed		
		Guinea Pigs	Rabbits	Rats
252	2.0	0/2	4/4	4/4
127	2.0	0/2	3/4	2/4
92	2.0	0/2	0/4	0/4
64	2.0	0/2	0/4	0/4
65	7.0	0/2	4/4	4/4
44.3	7.0	1/2	2/4	3/4
32.2	7.0	0/2	0/4	1/4
19.0	7.0	0/2	1/4	0/4
19.5	24.0	0/2	0/4	2/4
8.4	24.0	0/2	0/4	0/4
Total		1/20	14/40	16/40
Percentage		5.0	35.0	40.0

Table 12

The Fate of Animals Following Exposure to Fogs Formed by Dropping Di-sec-Amyl Sebacate Into an Inconel Tube Heated to the Temperature of 900°F (482°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Animals that Died /Number of Animals Exposed		
		Guinea Pigs	Rabbits	Rats
84	2.0	0/2	2/4	2/4
53	2.0	0/2	1/4	1/4
55	7.0	0/2	4/4	4/4
33.7	7.0	0/2	0/4	1/4
Total		0/8	7/16	8/16
Percentage		0.0	43.7	50.0

Table 13

The Fate of Animals Following Exposure to Fogs Formed by Dropping Di-sec-Amyl Sebacate Into an Inconel Tube Heated to Temperatures of 600°F (316°C) or 550°F (288°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Temperature of Inconel °F	Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Animals that Died /Number of Animals Exposed		
			Guinea Pigs	Rabbits	Rats
600	66	7.0	0/2	3/4	4/4
550	64	7.0	0/2	0/4	0/4
Total			0/4	3/8	4/8
Percentage			0.0	26.7	50.0

Table 14

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Di-2-Ethylhexyl Adipate Into an Inconel Tube Heated to the Temperature of 700°F (371°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed			
		Cats	Guinea Pigs	Rabbits	Rats
283.2	2.0	1/1	2/2	2/2	4/4
218.0	2.0	1/1	0/2	2/2	4/4
173.5	2.0	0/1	1/2	2/2	2/4
116.7	2.0	-	0/2	1/3	0/4
49.2	2.0	0/1	0/2	0/2	0/4
25.8	2.0	0/1	0/2	0/2	0/4
19.2	2.0	0/1	0/2	0/2	0/4
108.0	7.0	1/1	2/2	2/2	4/4
64.1	7.0	0/1	1/2	2/2	4/4
54.0	7.0	1/1	0/2	2/2	2/4
49.5	7.0	0/1	0/2	1/2	3/4
24.8	7.0	0/1	0/2	0/2	0/4
30.5	24.0	-	0/2	3/3	4/4
10.7	24.0	0/1	0/2	1/2	2/4
7.2	24.0	0/1	0/2	0/2	0/4
Total		4/13	6/30	18/32	29/60
Percentage		30.2	20.0	56.3	48.3

Table 15

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Di-2-Ethylhexyl Adipate Into an Inconel Tube Heated to the Temperature of 500°F (260°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed			
		Cats	Guinea Pigs	Rabbits	Rats
67.5	2.0	0/1	0/2	1/2	0/4
53.1	2.0	0/1	0/2	2/2	1/4
97.4	7.0	1/1	1/2	2/2	4/4
59.0	7.0	1/1	0/2	2/2	1/4
24.5	7.0	0/1	0/2	0/1	0/4
Total		2/5	1/10	7/9	6/20
Percentage		40.0	10.0	77.8	30.0

Table 16

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Di-2-Ethylhexyl Adipate Into an Inconel Tube Heated to the Temperature of 400°F (204°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed			
		Cats	Guinea Pigs	Rabbits	Rats
172.5	2.0	0/1	(1) 1/2	0/2	0/4
152.1	7.0	0/1	0/2	0/2	0/4
94.3	7.0	0/1	0/2	0/2	0/4
81.9	7.0	0/1	0/2	0/2	0/4
Total		0/4	1/8	0/8	0/16
Percentage		0.0	12.5	0.0	0.0

(1) This death resulted from pneumonia, unrelated to exposure.

Table 17

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Unused WS-2211 Into an Inconel Tube Heated to the Temperature of 700°F (371°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed			
		Cats	Guinea Pigs	Rabbits	Rats
207.0	2.0	1/1	0/2	2/2	2/2
129.2	2.0	-	0/2	3/3	2/2
86.7	2.0	-	0/2	1/2	1/2
60.0	2.0	-	0/2	0/3	0/4
45.0	2.0	0/1	0/2	1/2	0/4
61.4	7.0	-	1/2	3/3	4/4
43.1	7.0	-	0/2	3/3	4/4
30.2	7.0	-	0/2	2/3	3/4
19.5	7.0	-	0/2	0/3	1/4
15.0	7.0	-	0/2	1/3	0/4
17.9	24.0	-	0/2	3/3	2/4
7.4	24.0	0/1	0/2	0/2	0/4
Total		1/3	1/24	19/32	19/42
Percentage		33.3	4.2	59.4	45.2

Table 18

The Fate of Animals Following Exposure to the Fogs Formed by Dropping Unused WS-2211 Into an Inconel Tube Heated to Temperatures of 400°F (204°C) or 550°F (288°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Temperature of Inconel °F	Rate of Dropping Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed			
			Cats	Guinea Pigs	Rabbits	Rats
550	32.9	7.0	-	0/2	1/3	1/4
400	83.6	7.0	0/1	0/2	0/2	0/4
400	67.8	7.0	-	0/2	1/3	0/4
400	49.8	7.0	-	0/2	0/3	1/4
400	41.7	7.0	0/1	0/2	0/2	0/4
400	25.0	7.0	-	0/2	0/3	0/4
Total			0/2	0/10	1/13	1/20
Percentage			0.0	0.0	7.7	5.0

Table 17

The Fate of Animals Following Exposure to the Fogs Formed by Dropping PRL-3039 Into an Inconel Tube Heated to the Temperature of 700°F (371°C)

(Rate of Passage of Air Through Furnace: 31.8 l/min)

Rate of Dropping of Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed		
		Guinea Pigs	Rabbits	Rats
195	2	1/2	4/4	4/4
114	2	0/2	2/4	1/4
91	2	0/1	2/4	0/4
62	2	0/2	0/4	0/4
65	7	0/2	4/4	3/4
45	7	0/2	2/4	0/4
30.1	7	0/2	1/4	0/4
19.4	7	0/2	0/4	0/4
19.3	24	0/2	2/4	0/4
7.3	24	0/2	2/4	0/4
Total		1/19	19/40	8/40
Percentage		5.3	47.5	20.0

Table 20

The Fate of Animals Following Exposure to the Fogs Formed by Dropping PRL-3039 Into an Inconel Tube Heated to the Temperature of 900°F (482°C)

Rate of Dropping of Material Upon Inconel mg/min	Duration of Exposure Hours	Number of Fatalities/Number of Animals Exposed		
		Guinea Pigs	Rabbits	Rats
92	2	0/2	2/4	2/4
63	2	0/2	0/4	0/4
47	7	0/2	3/4	4/4
31.2	7	0/2	2/4	1/4
Total		0/8	7/16	7/16
Percentage		0.0	43.8	43.8

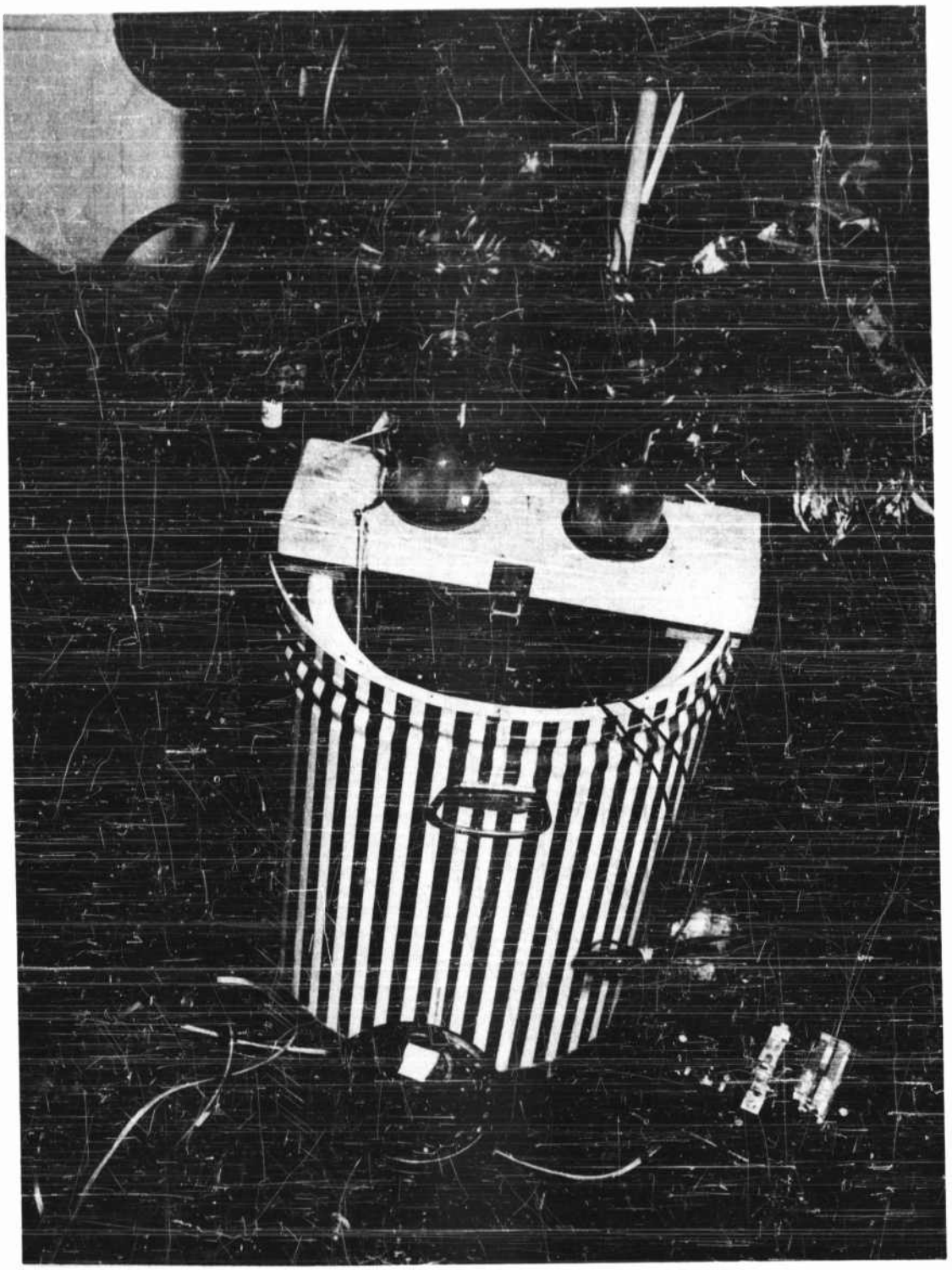


Figure 1. Bubbling Towers for Volatilization of Vapors of the Aliphatic Esters

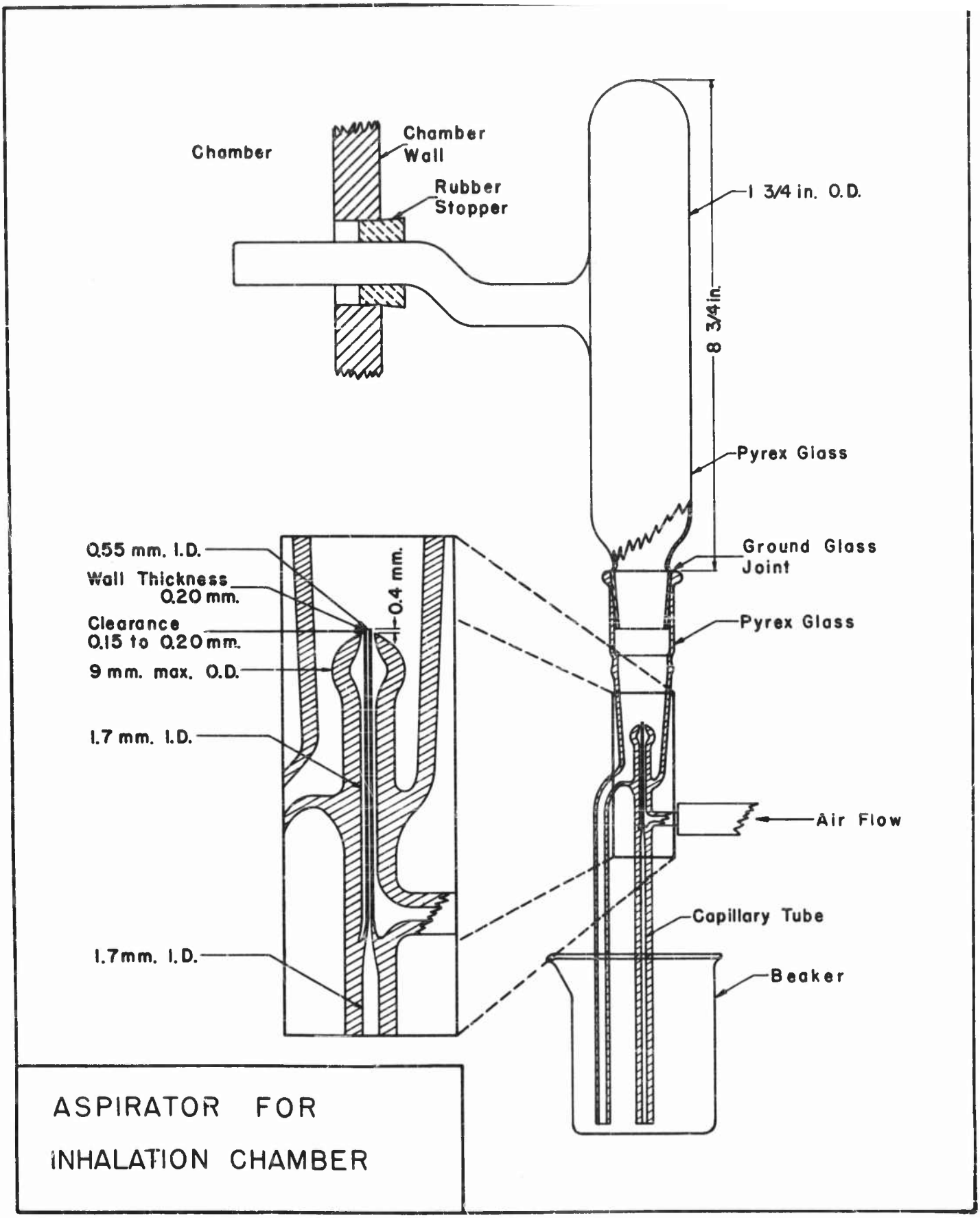


Figure 2. Sketch of Kettering Type of Aspirator for Generation of a Mist

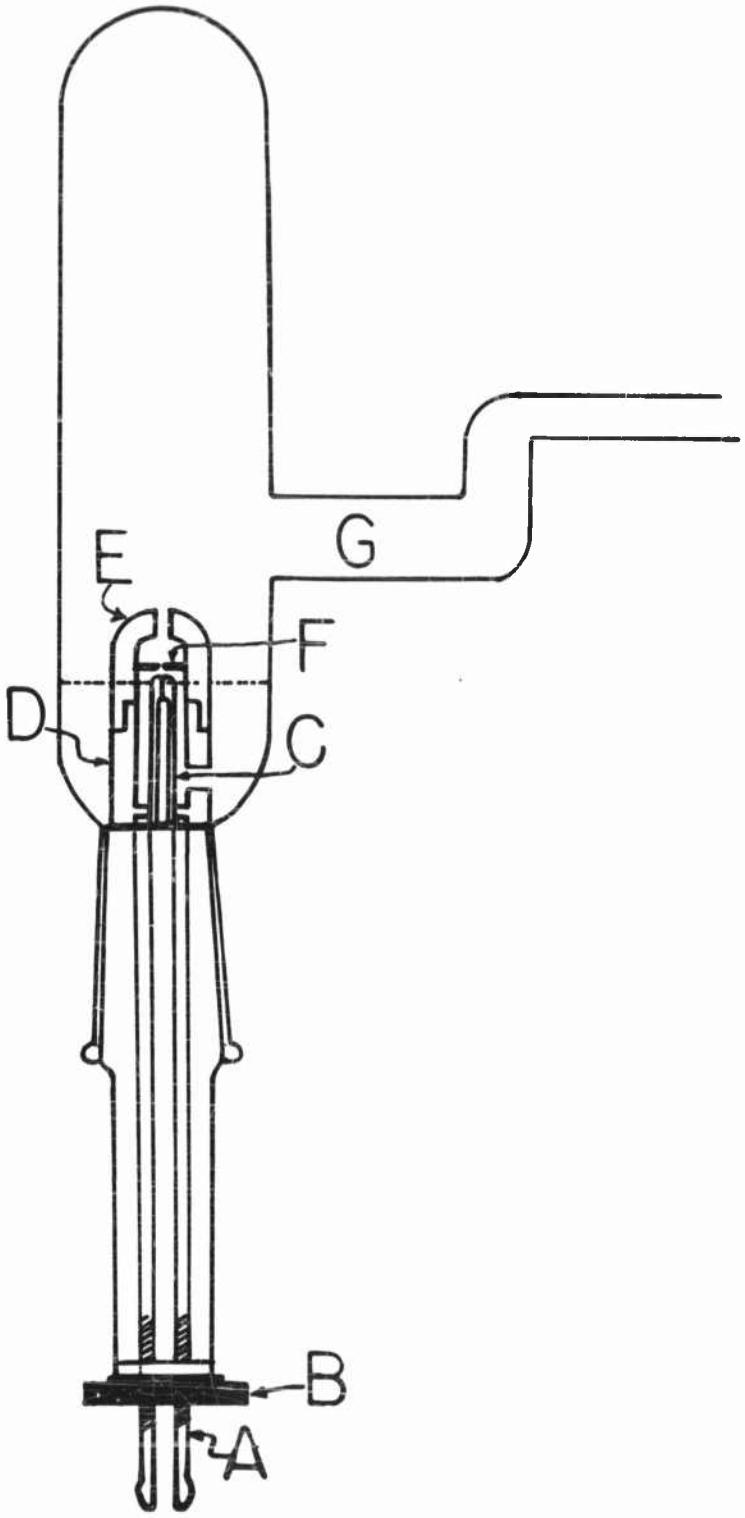


Figure 3. Sketch of Lundegårdh Type of Aspirator for Generation of a Mist in the Air

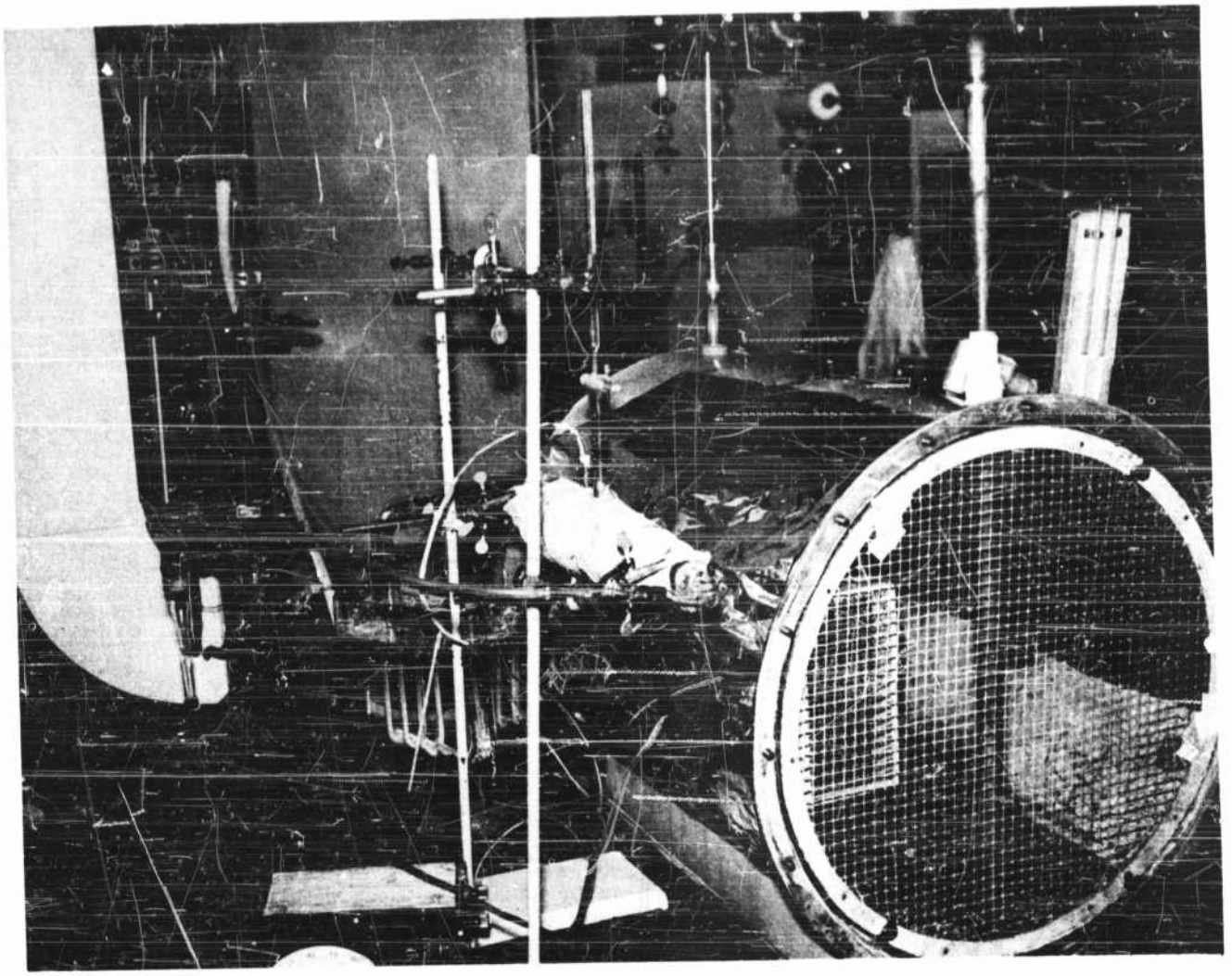


Figure 4. Equipment for the Generation of Decomposition Products at High Temperatures and Front View of 223-Liter Chamber

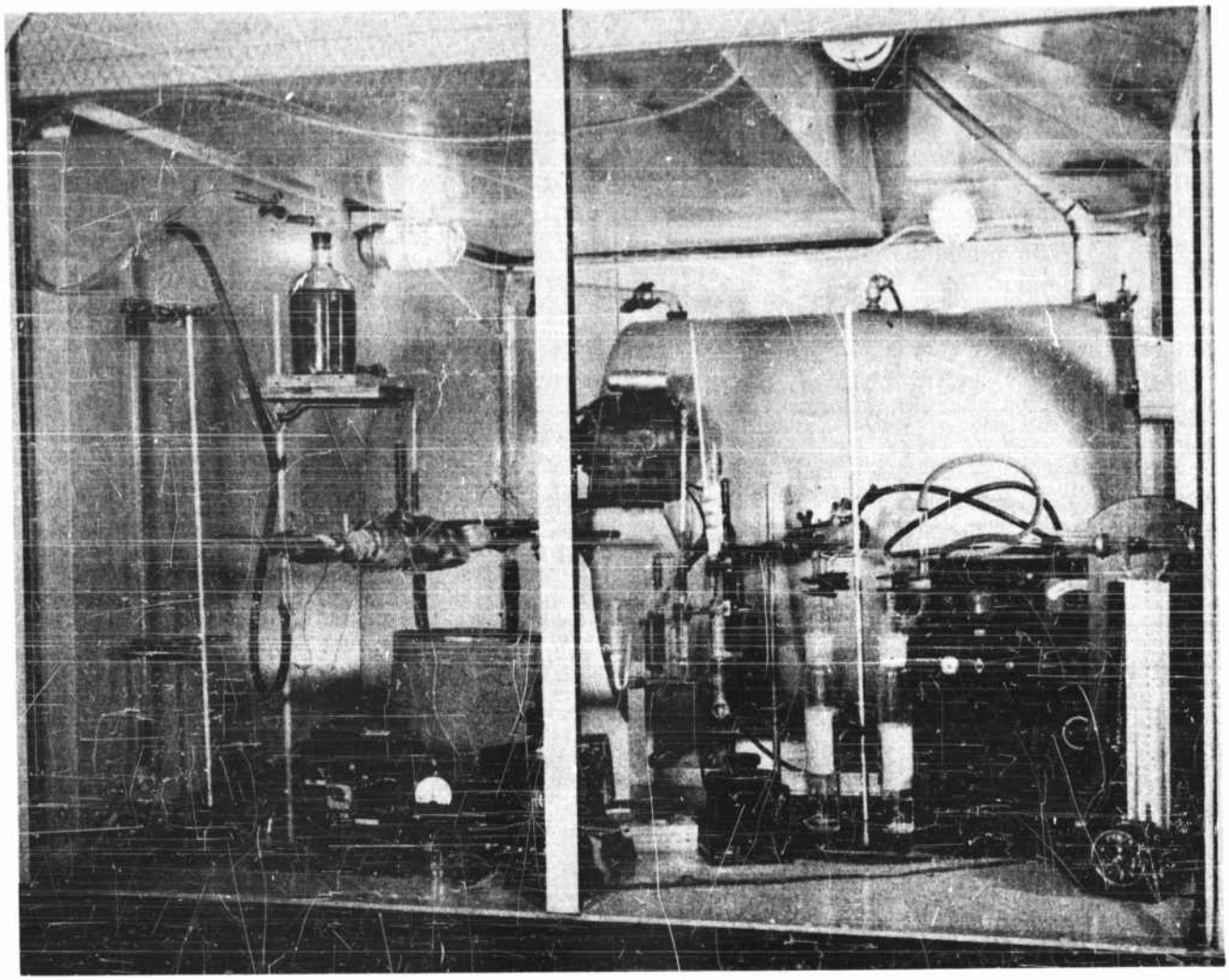


Figure 5. Equipment for the Generation of Decomposition Products at High Temperatures, Sampling Equipment and Side View of 800-liter Chamber

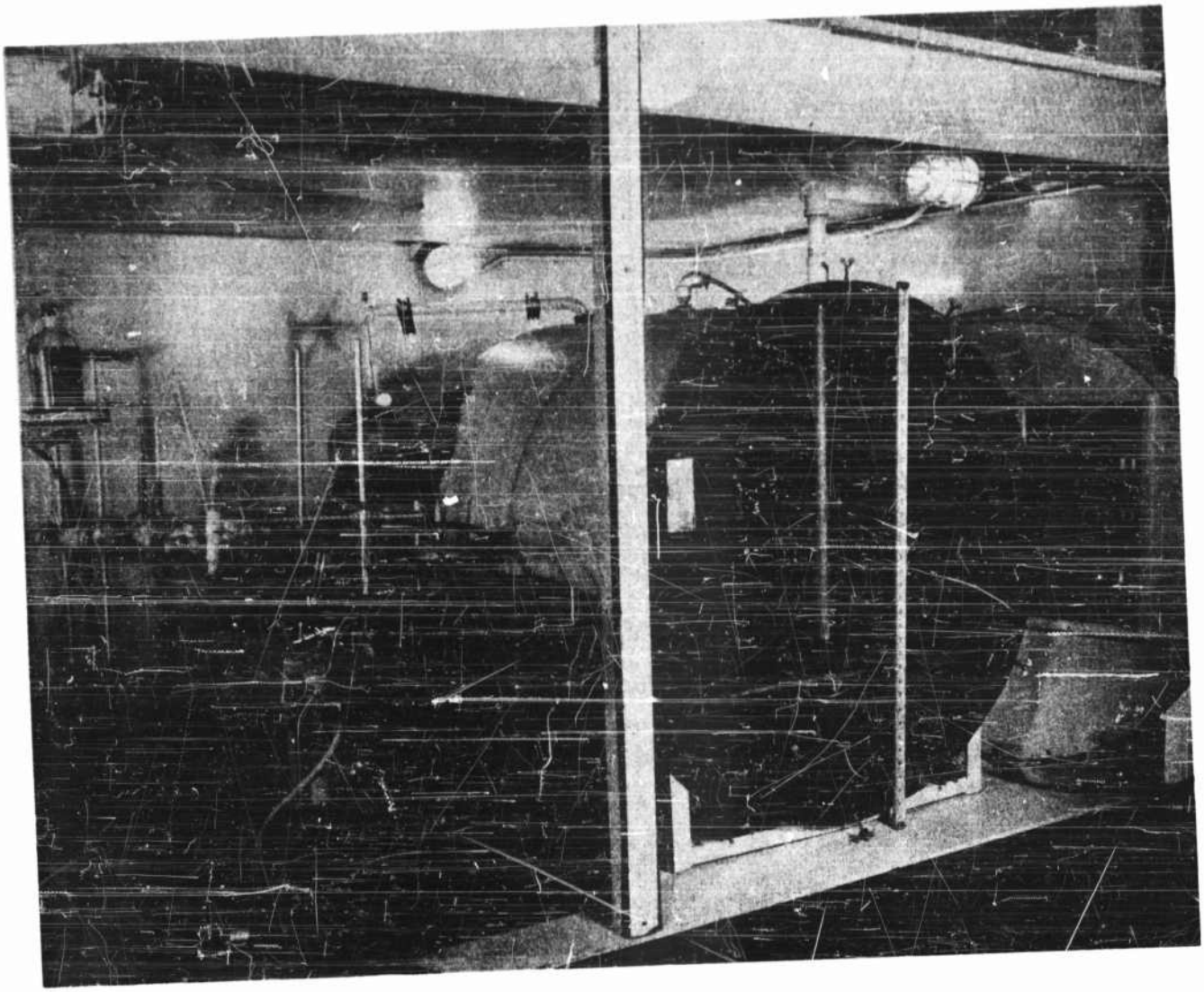


Figure 6. Front View of 800-Liter Chamber

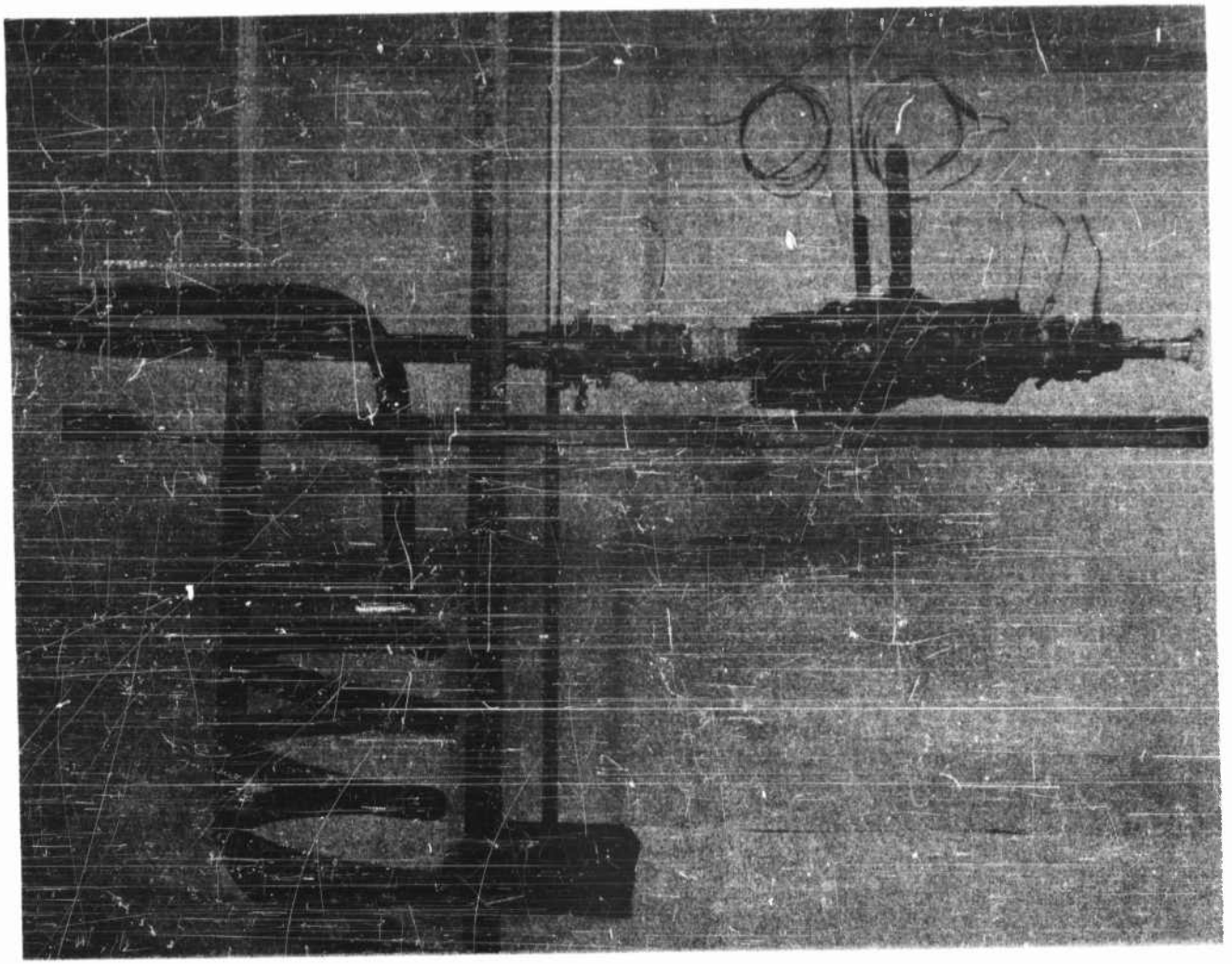


Figure 7. Furnace for Thermal Decomposition and Condenser Coils

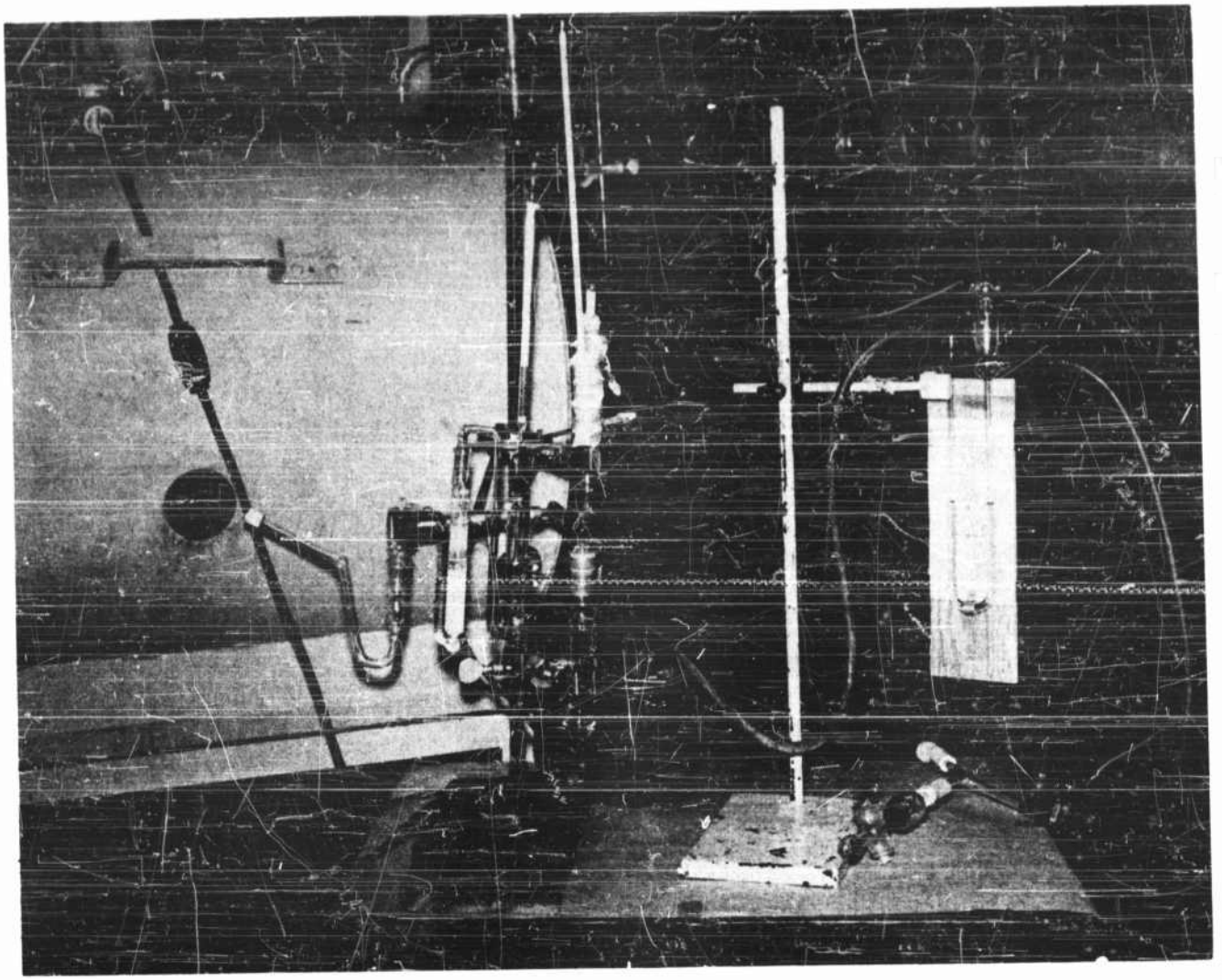


Figure 8. Absorption Train for Collection of Carbon Monoxide from Air

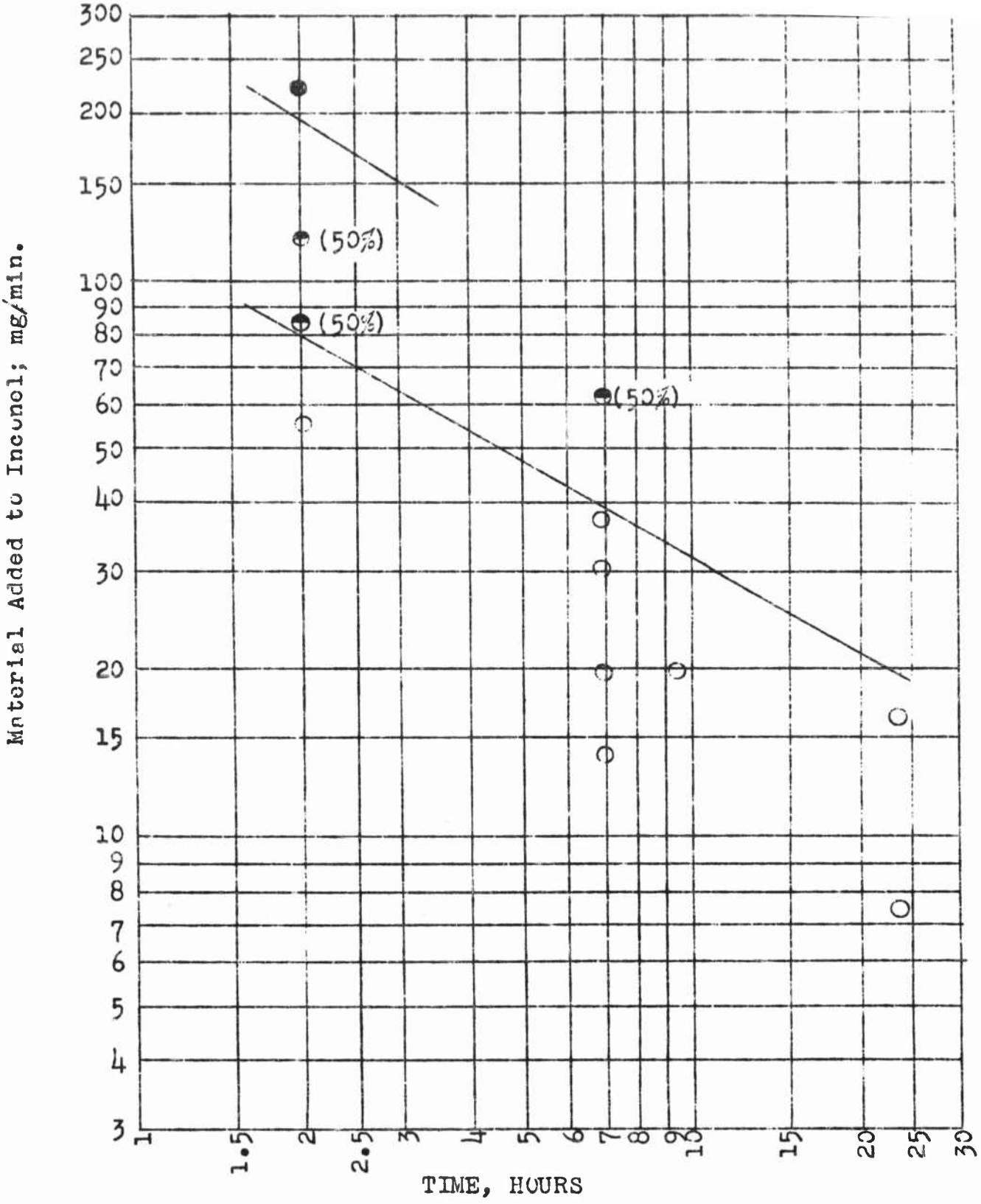


Figure 9. Fatalities Among Guinea-Pigs Following the Inhalation of the Fog Formed by Dropping Di-2-Ethylhexyl Sebacate Into an Inconel Tube to 700°F

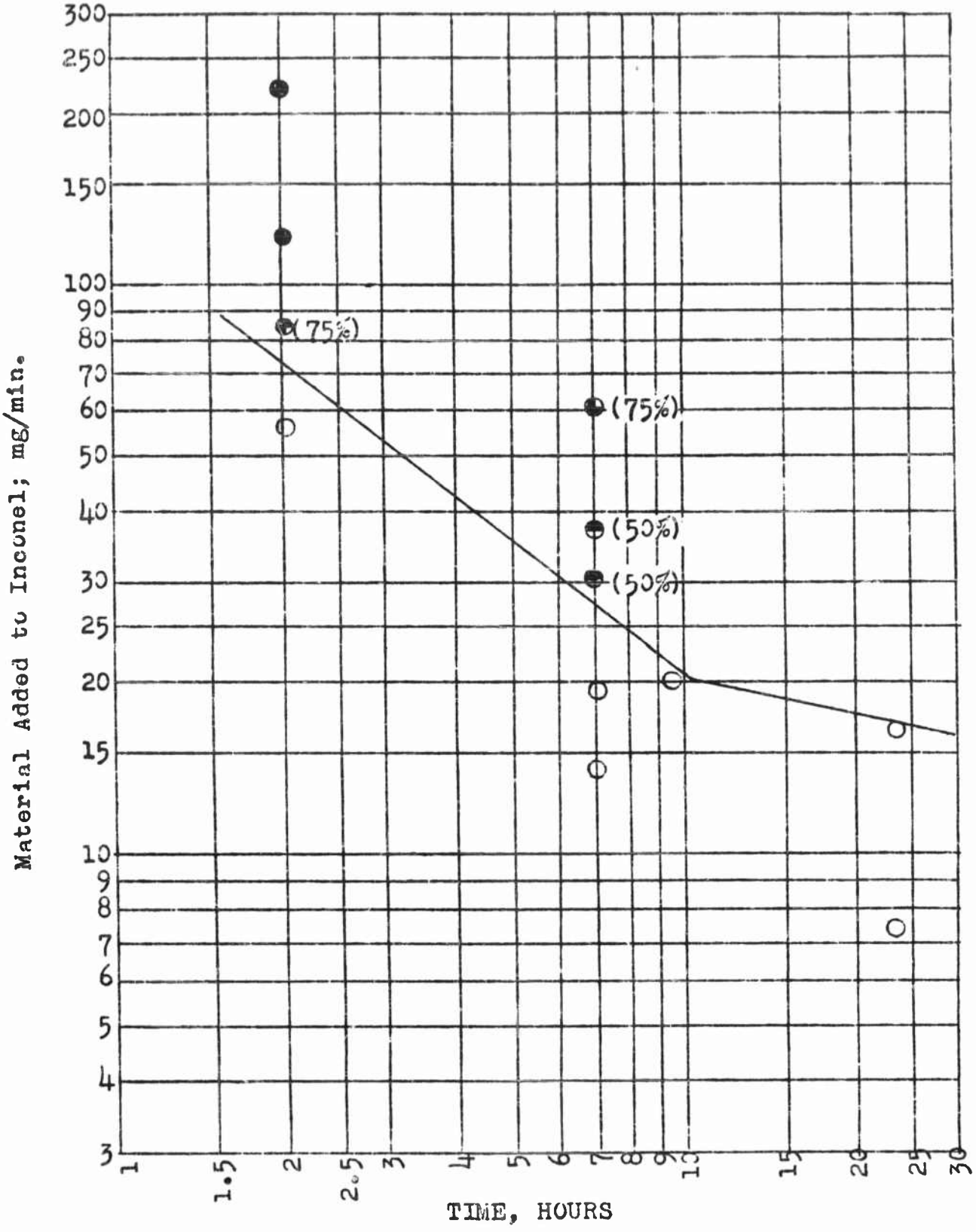


Figure 10. Fatalities Among Rabbits Following the Inhalation of the Fog Formed by Dropping Di-2-Ethylhexyl Sebacate Into an Inconel Tube Heated to 700°F

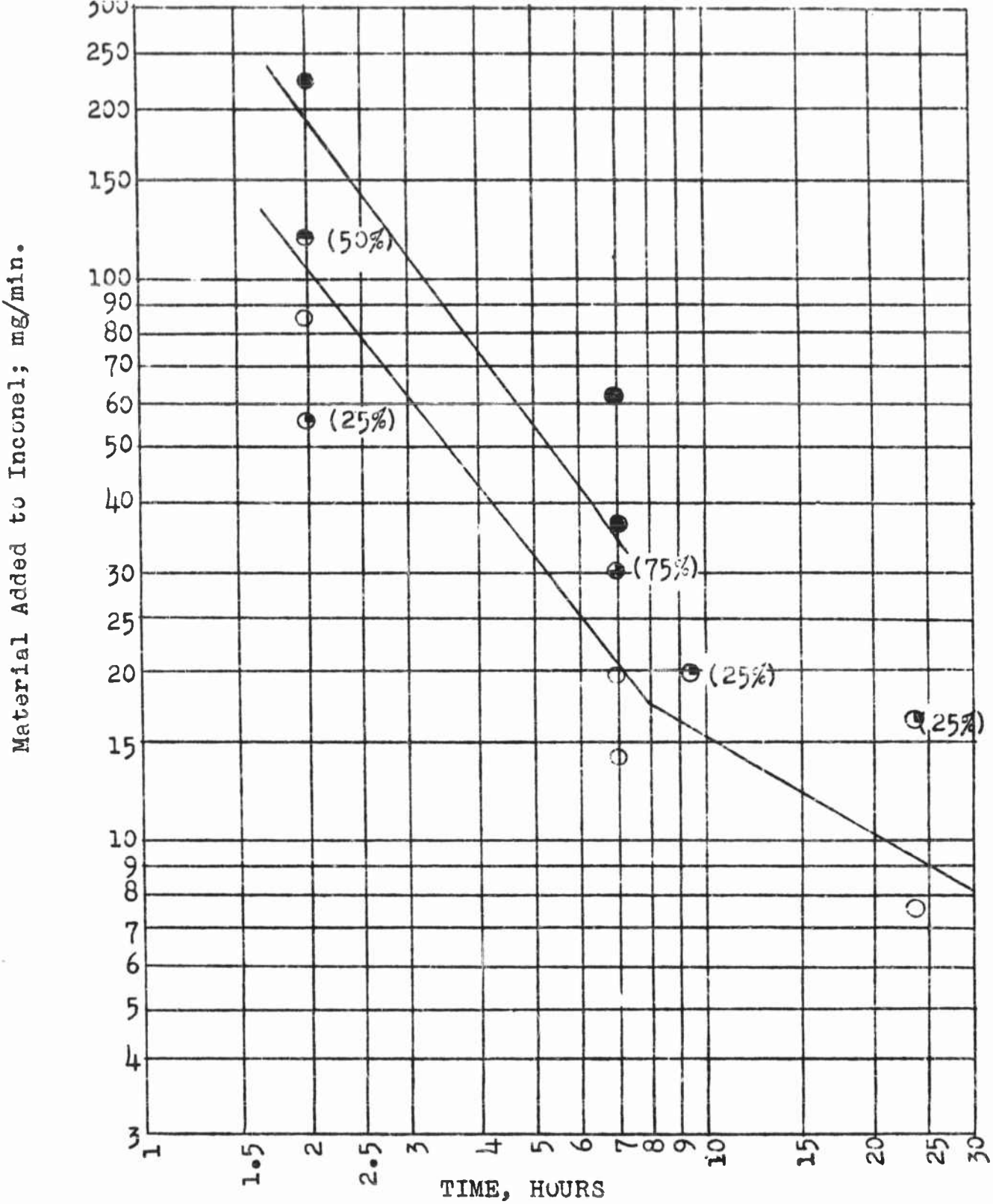


Figure 11. Fatalaties Among Rats Following the Inhalation of the Fog Formed by Dropping Di-2-Ethylhexyl Sebacate Into an Inconel Tube Heated to 700°F

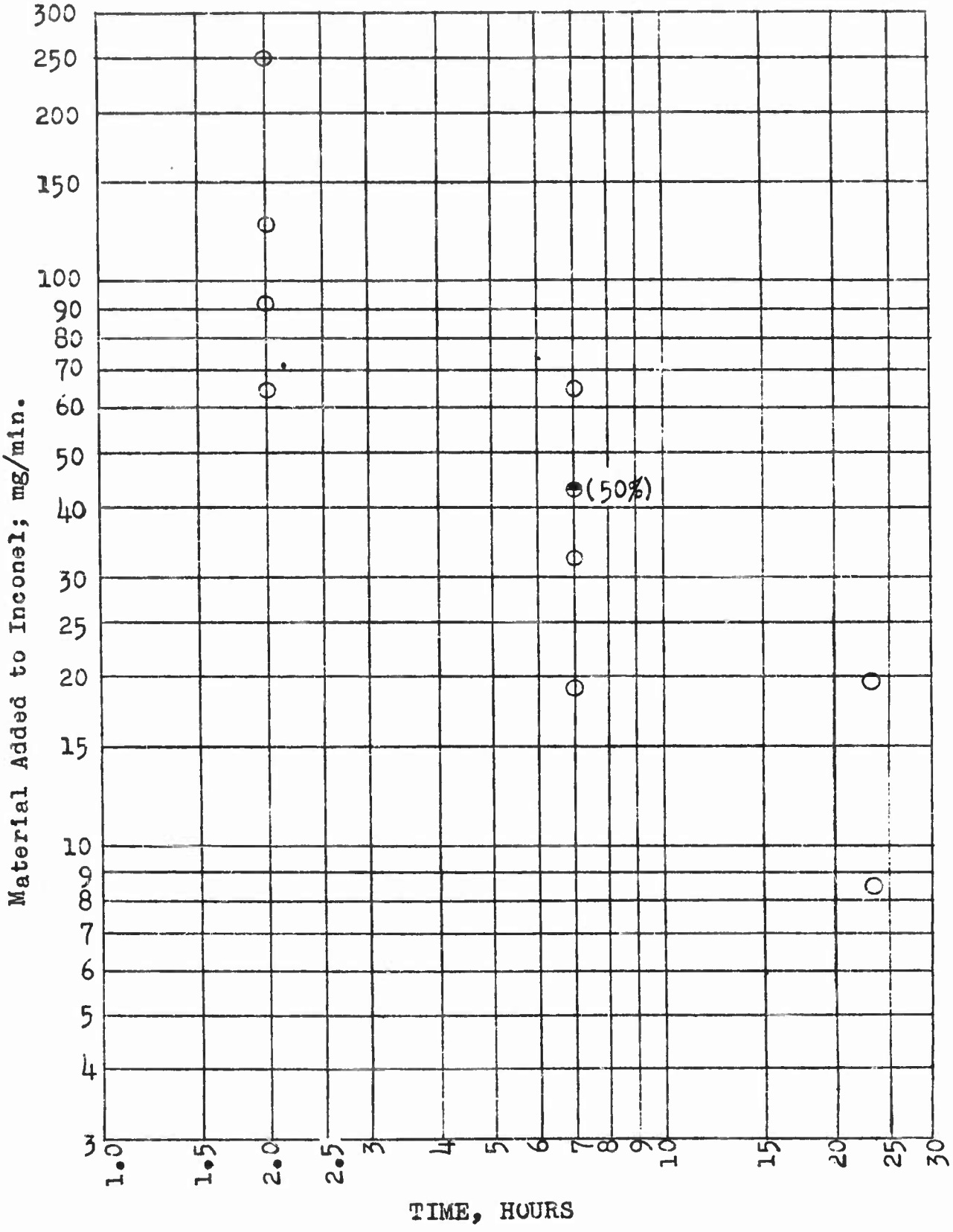


Figure 12. Fatalaties Among Guinea Pigs Following the Inhalation of the Fog Formed by Dropping Secondary Amyl Sebacate Into an Inconel Tube Heated to 700°F

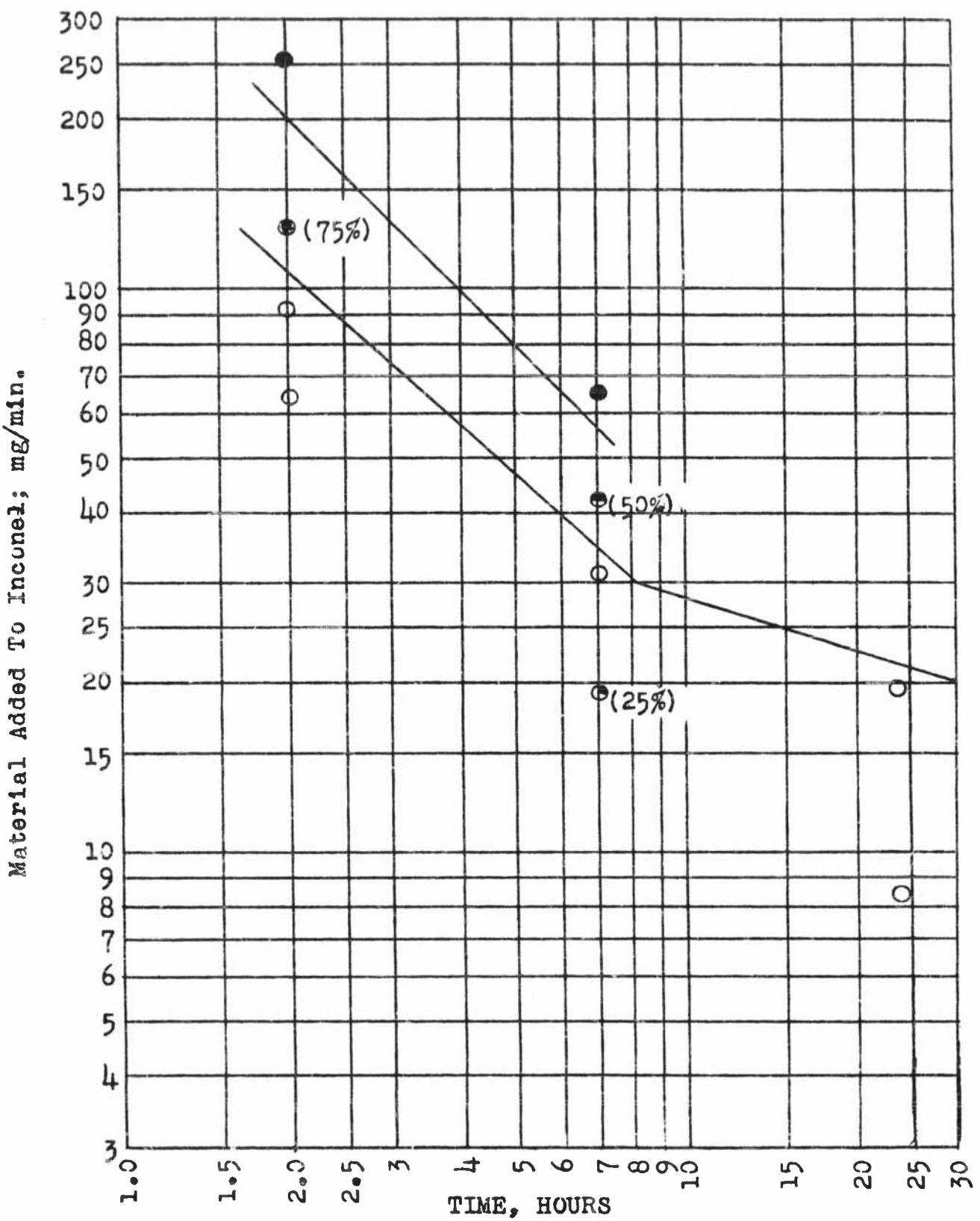


Figure 13. Fatalities Among Rabbits Following the Inhalation of the Fog Formed by Dropping Secondary Amyl Sebacate Into an Inconel Tube Heated to 700°F

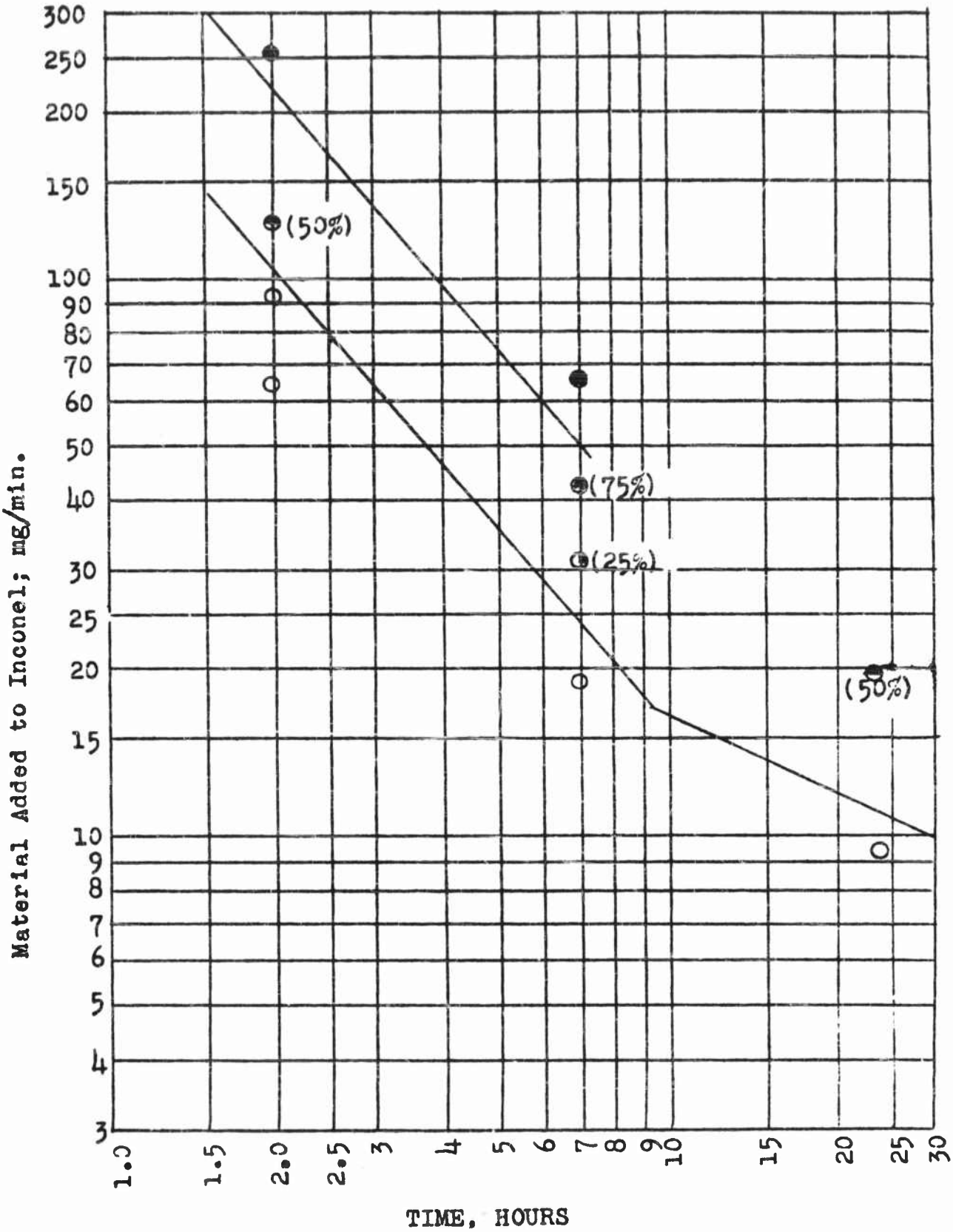


Figure 14. Fatalities Among Rats Following the Inhalation of the Fog Formed by Dropping Secondary Amyl Sebacate Into an Inconel Tube Heated to 700°F

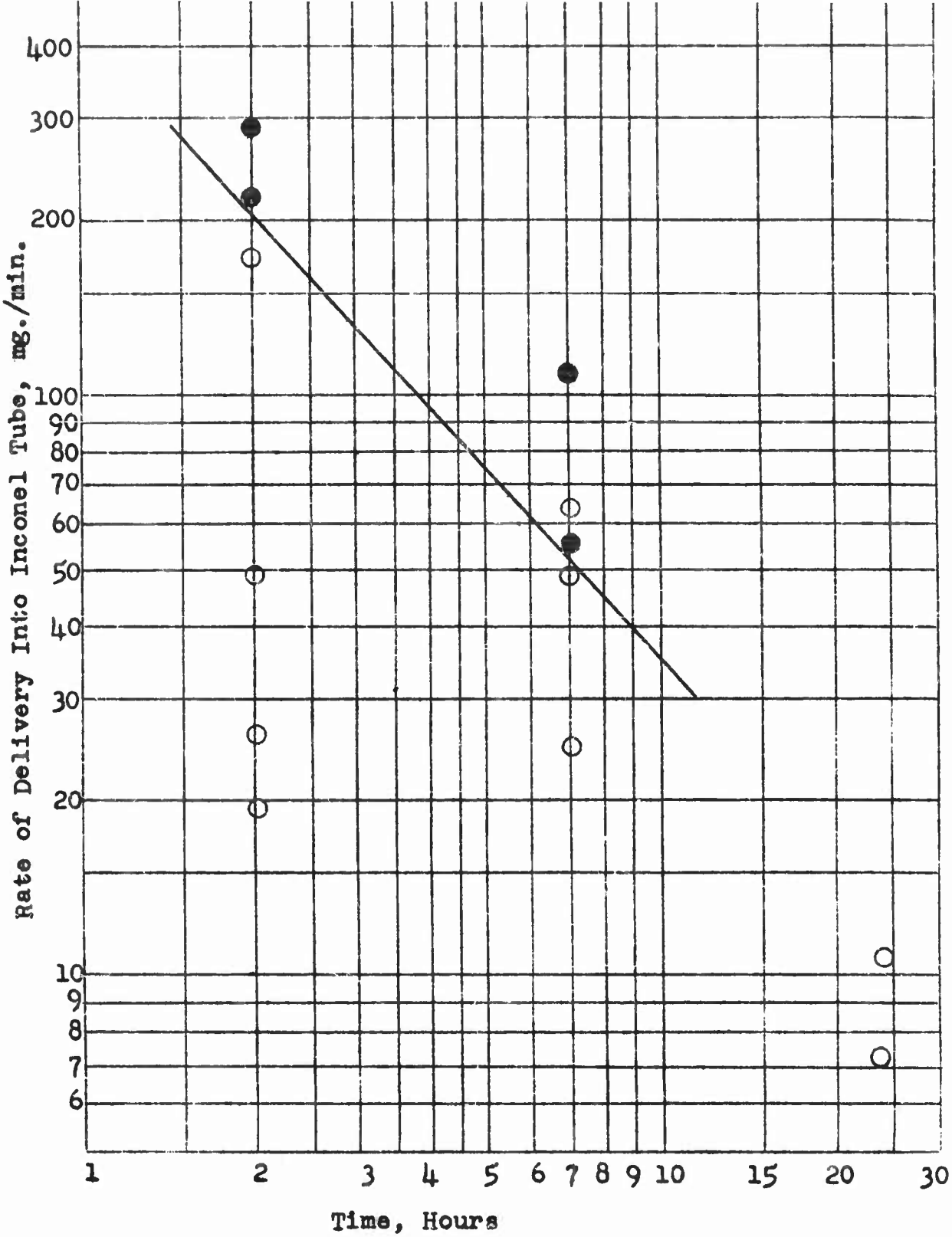


Figure 15. Fatalities Among Cats Following the Inhalation of the Fog Formed by Dropping Di-2-Ethylhexyl Adipate Into an Inconel Tube Heated to 700°F

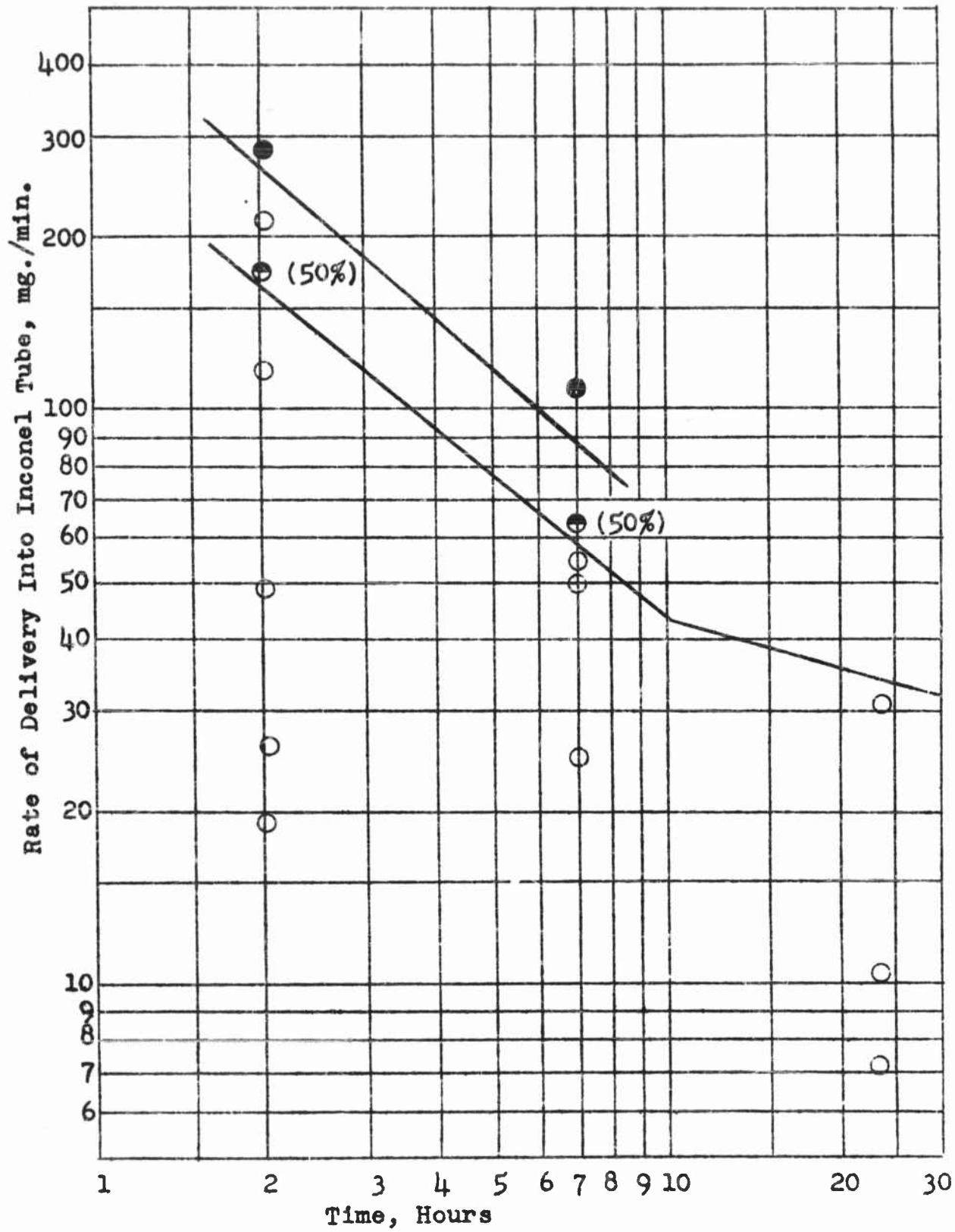


Figure 16. Fatalities Among Guinea Pigs Following the Inhalation of the Fog Formed by Dropping Di-2-Ethylhexyl Adipate Into an Inconel Tube Heated to 700°F

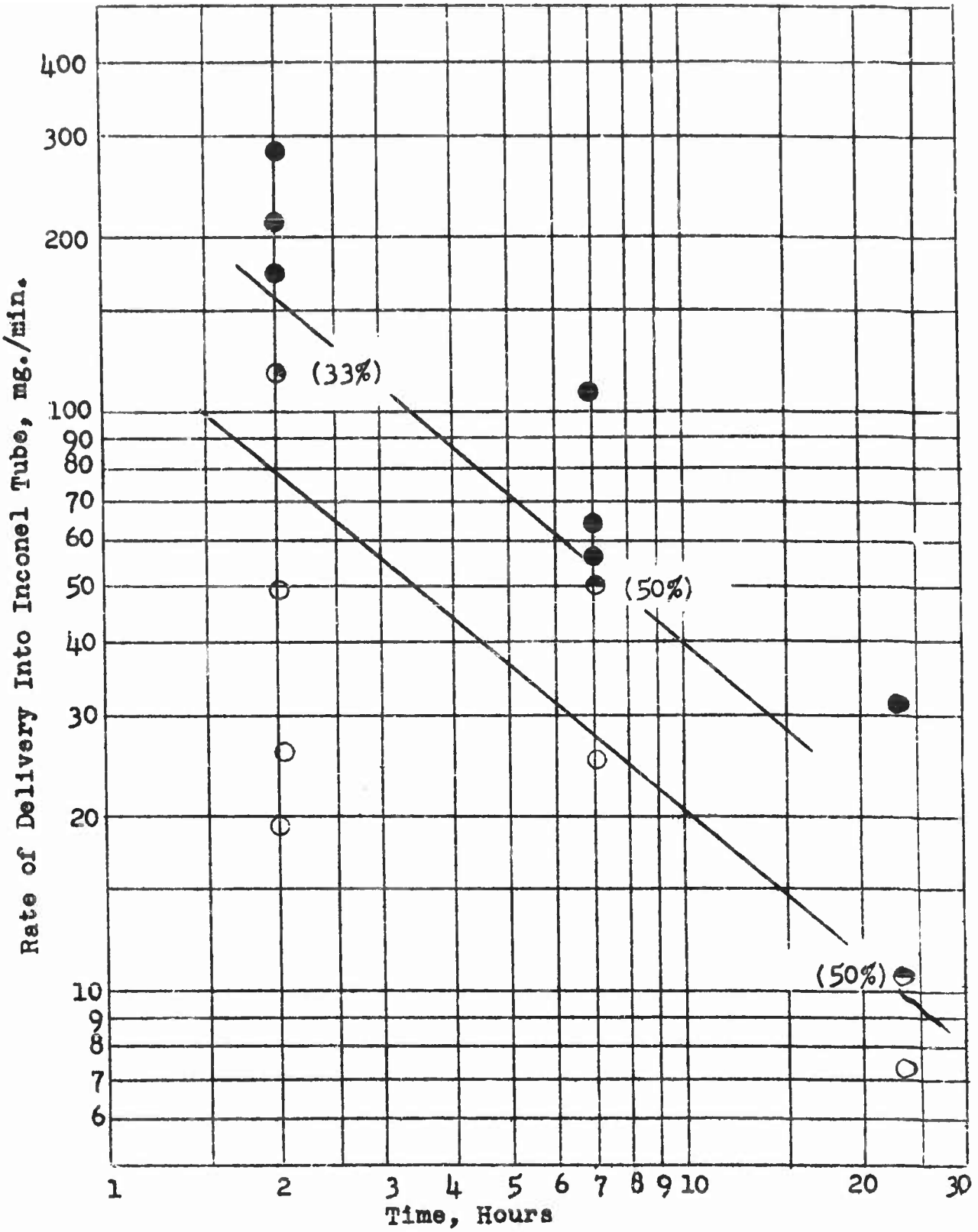


Figure 17. Fatalities Among Rabbits Following the Inhalation of the Fog Formed by Dropping Di-2-Ethylhexyl Adipate Into an Inconel Tube Heated to 700°F

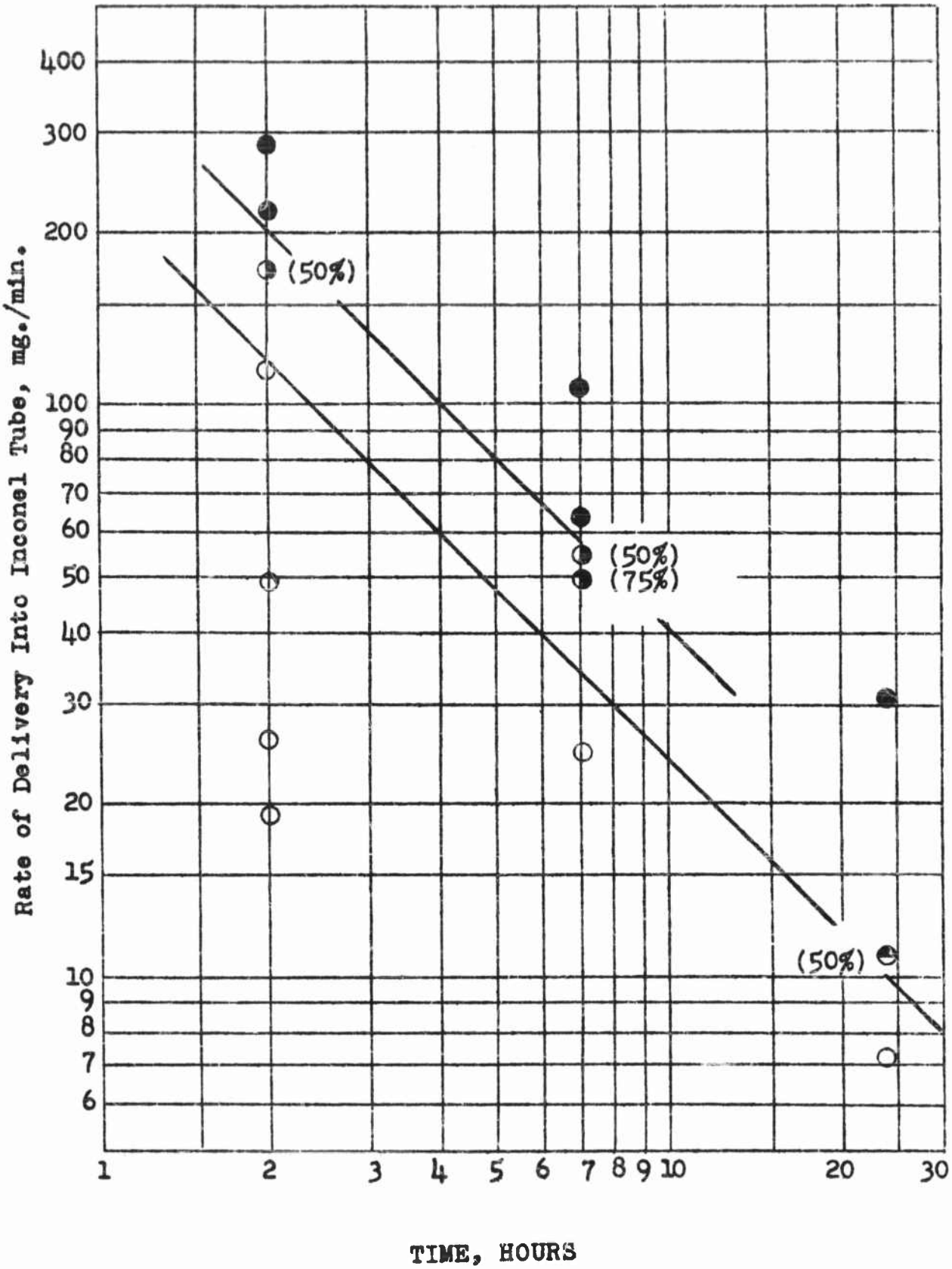


Figure 18. Fatalities Among Rats Following the Inhalation of the Fog Formed by Dropping Di-2-Ethylhexyl Adipate Into an Inconel Tube Heated to 700°F

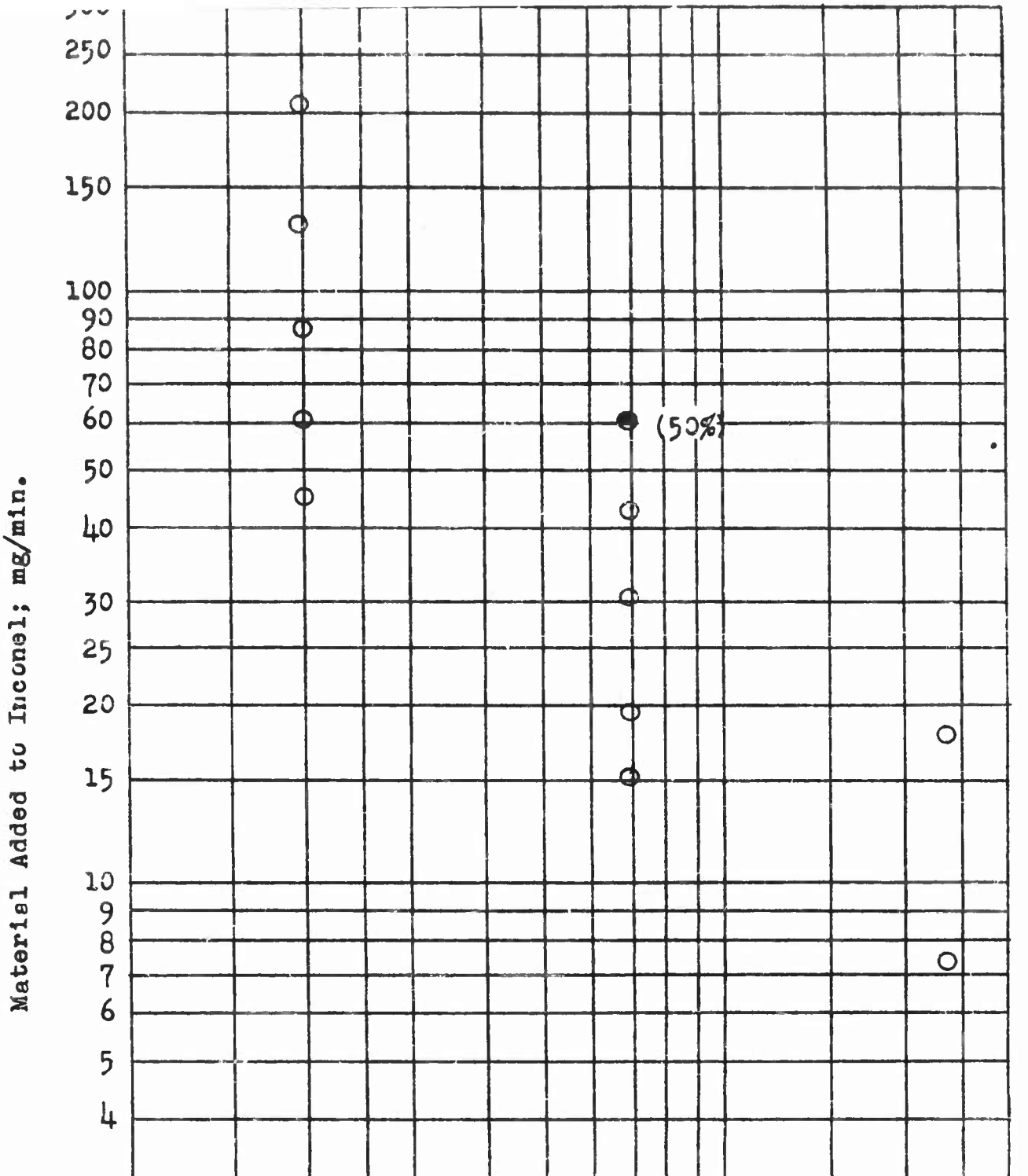


Figure 19. Fatalities Among Guinea-Pigs Following the Inhalation of the Fog Formed by Dropping WS-2211 Into an Inconel Tube Heated to 700°F

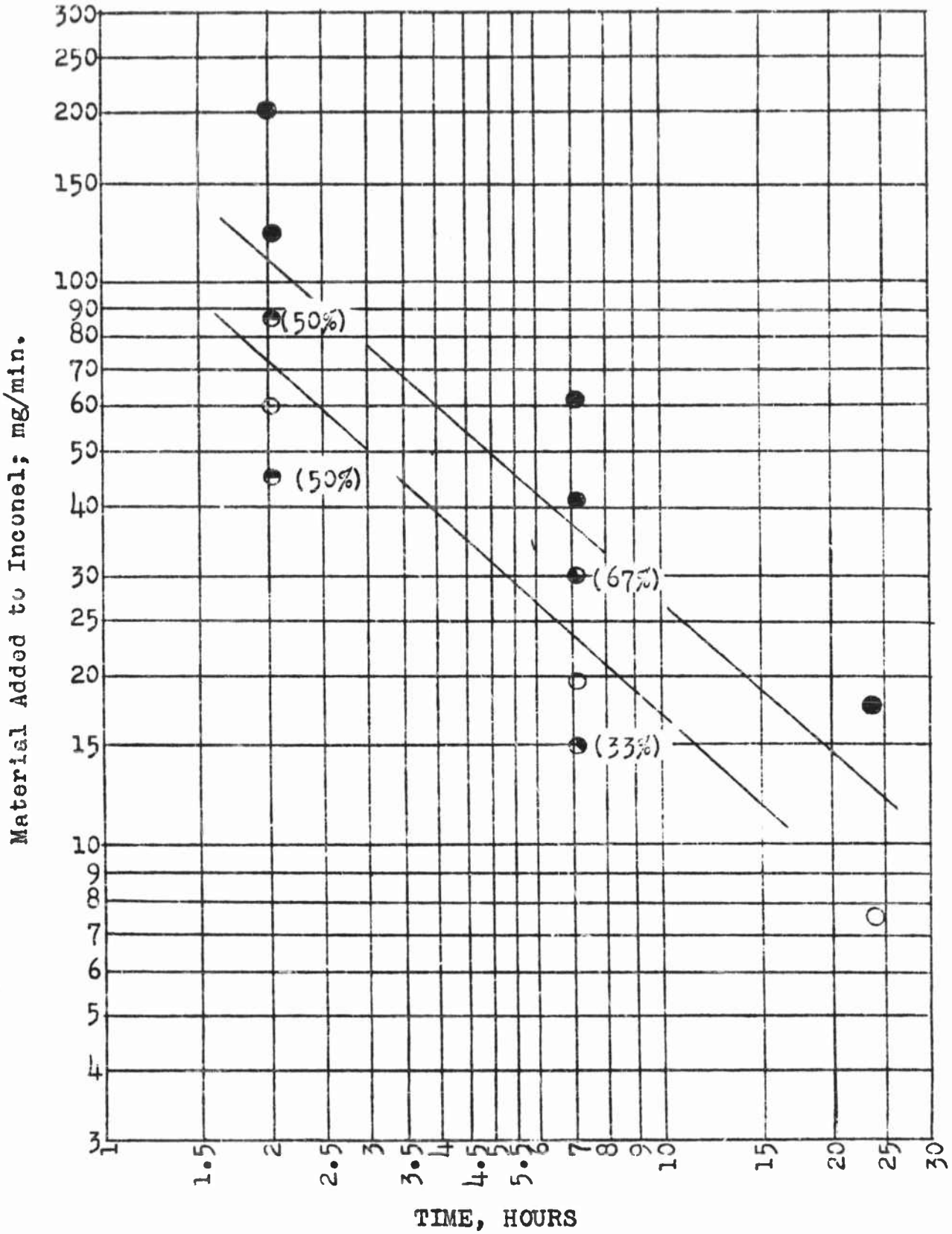


Figure 20. Fatalities Among Rabbits Following the Inhalation of the Fog Formed by Dropping WS-2211 Into an Inconel Tube Heated to 700°F

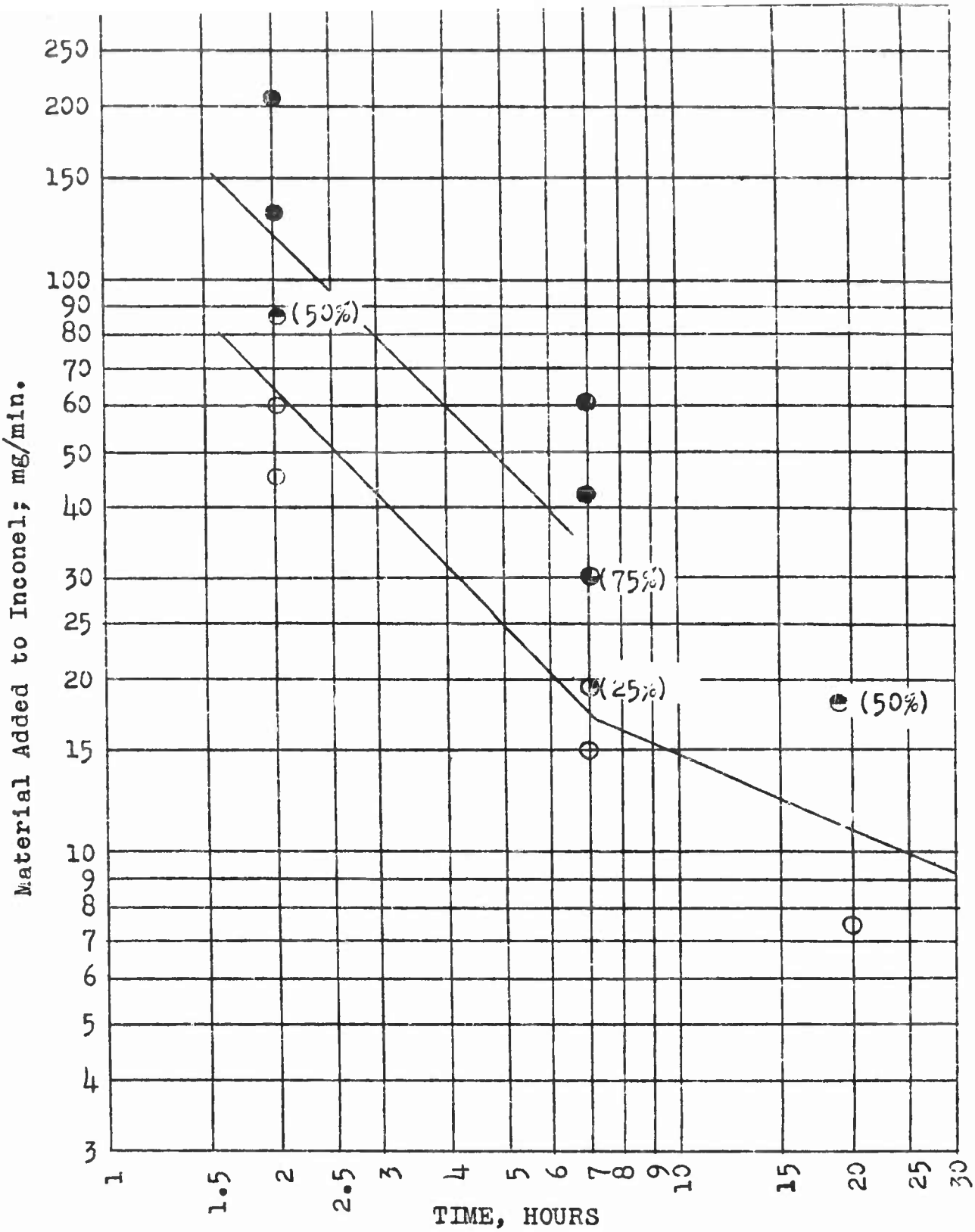


Figure 21. Fatalaties Among Rats Following the Inhalation of the Fog Formed by Dropping WS-2211 Into an Inconel Tube Heated to 700°F

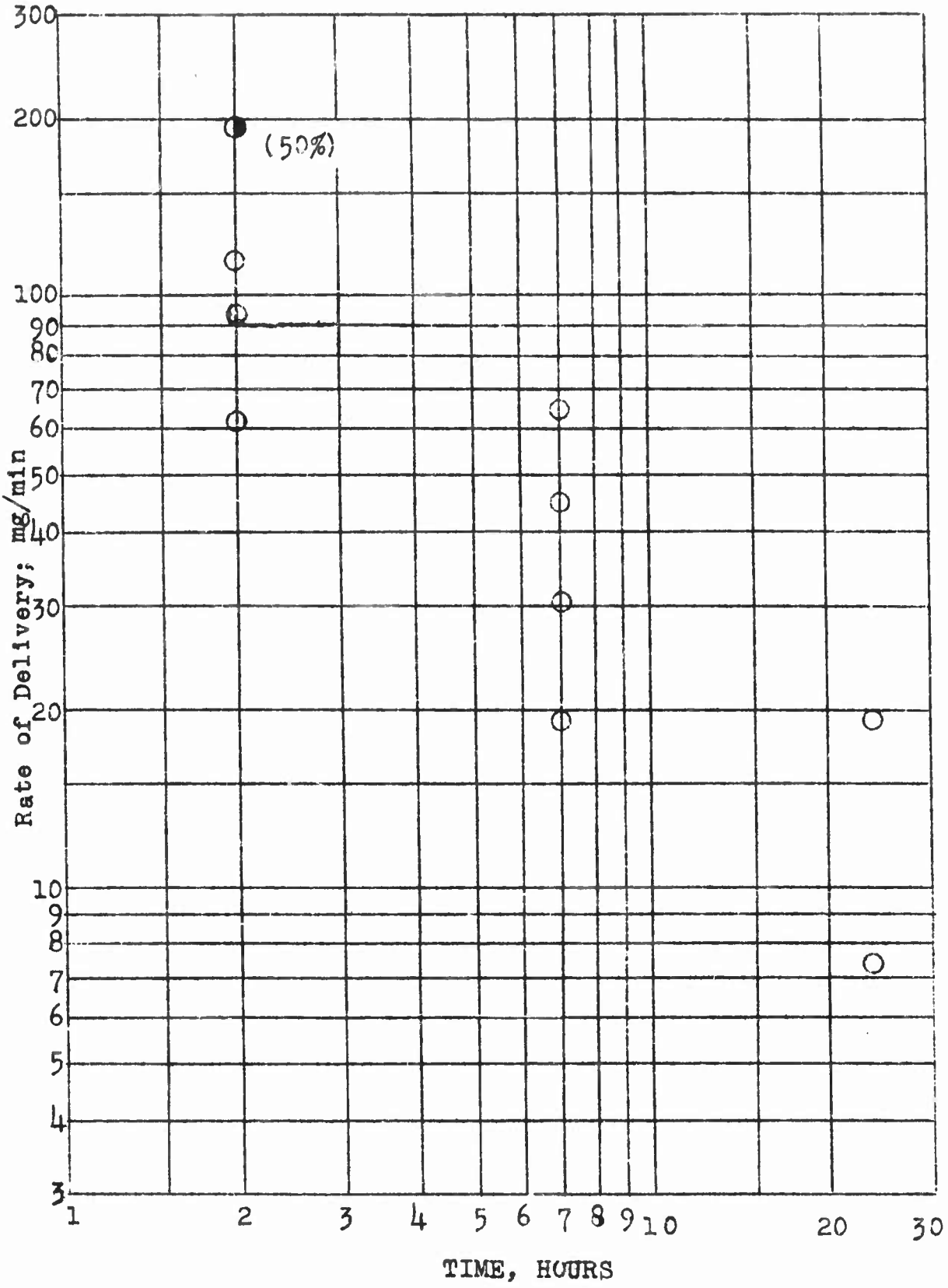


Figure 22. Fatalaties Among Guinea Pigs Following the Inhalation of the Fog Formed by Delivering PRL-3039 Into an Inconel Tube Heated to 700°F

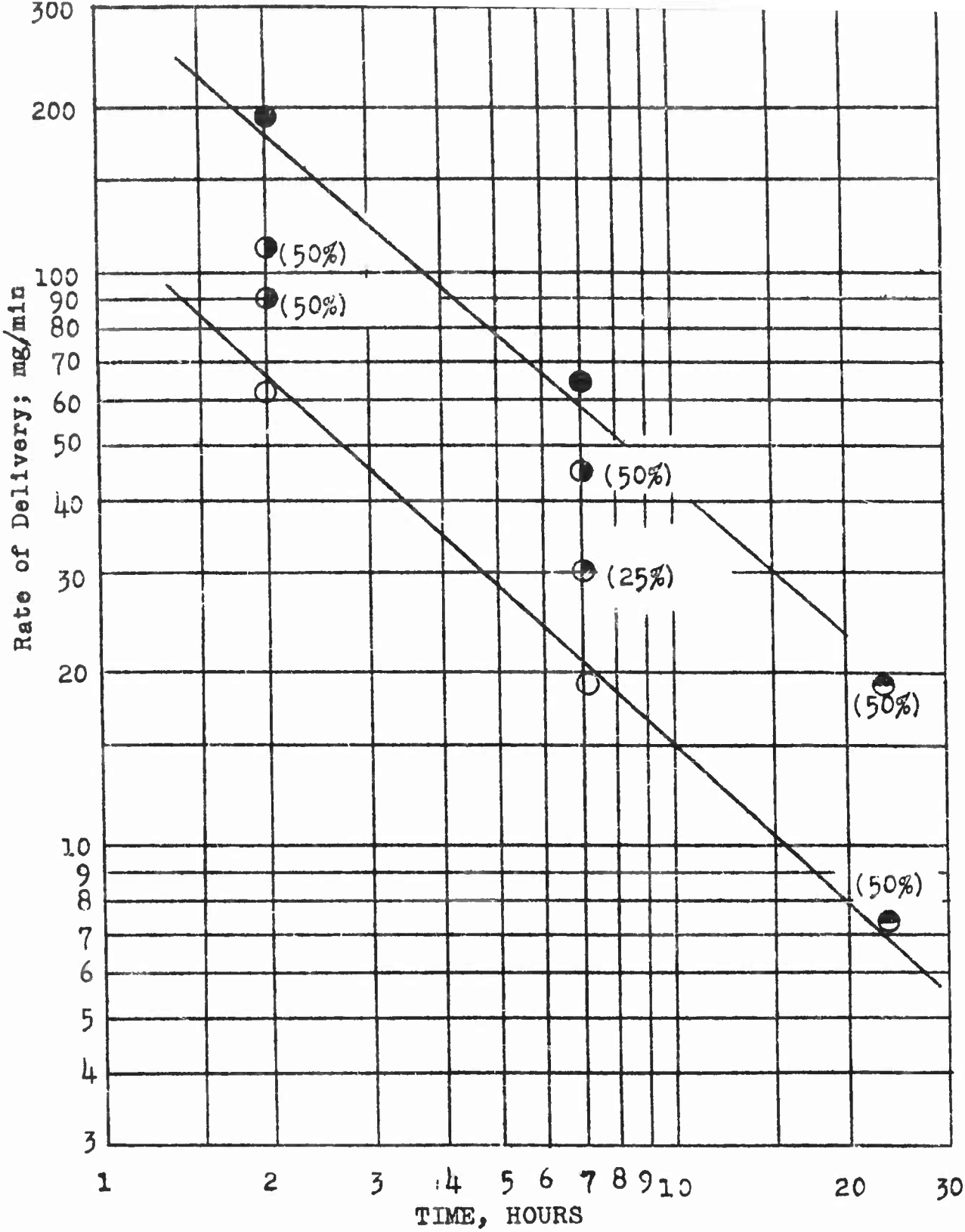


Figure 23. Fatalaties Among Rabbits Following the Inhalation of the Fog Formed by Dropping PRL-3039 Into an Inconel Tube Heated to 700°F

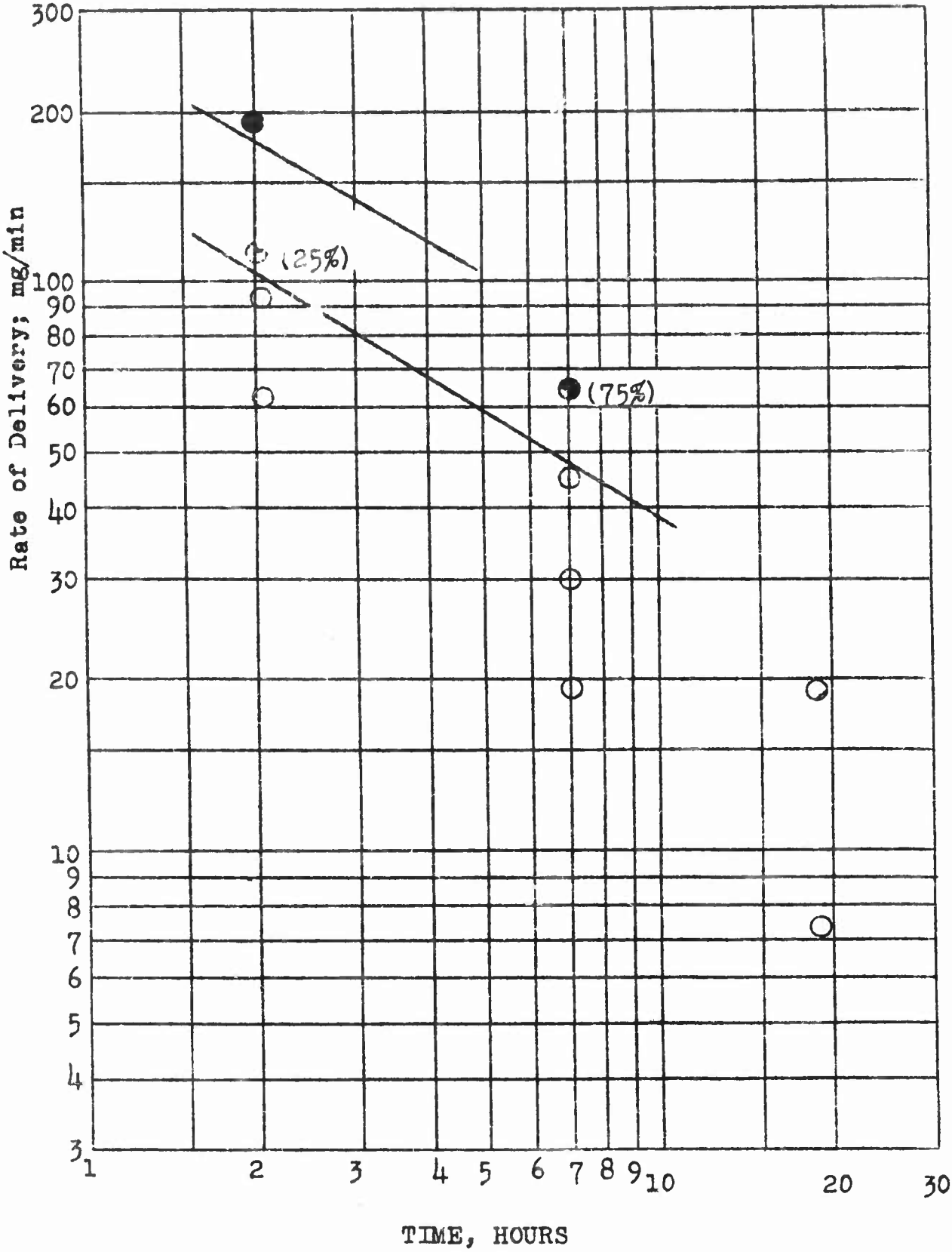


Figure 24. Fatalities Among Rats Following the Inhalation of the Fog Formed by Delivering PRL-3039 Into an Inconel Tube Heated to 700°F

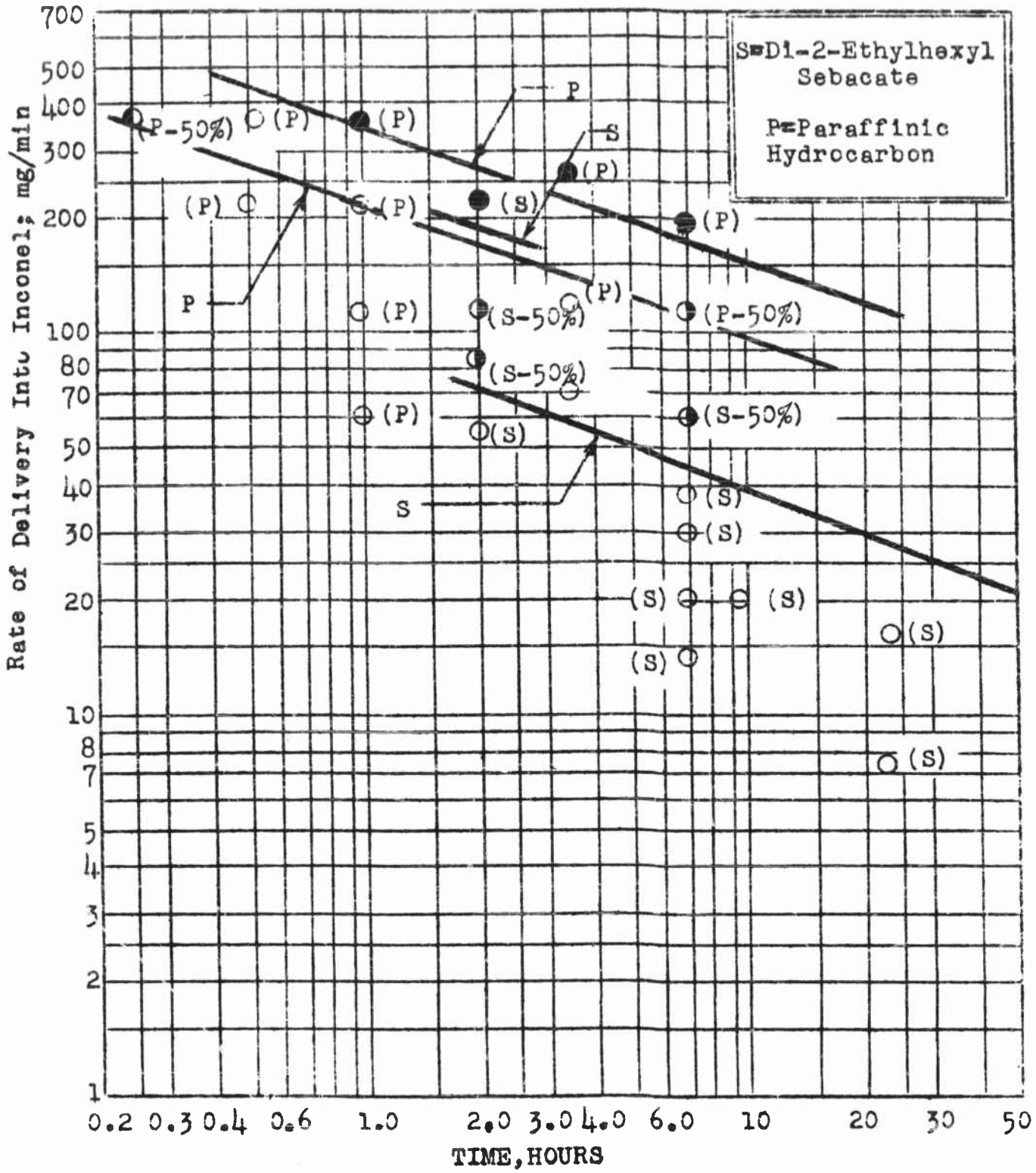


Figure 25. Fatalities Among Guinea Pigs Following the Inhalation of the Fog Formed by Dropping Either a Paraffinic Hydrocarbon or Di-2-Ethylhexyl Sebacate Into an Inconel Tube Heated to 800° or to 700°F, Respectively

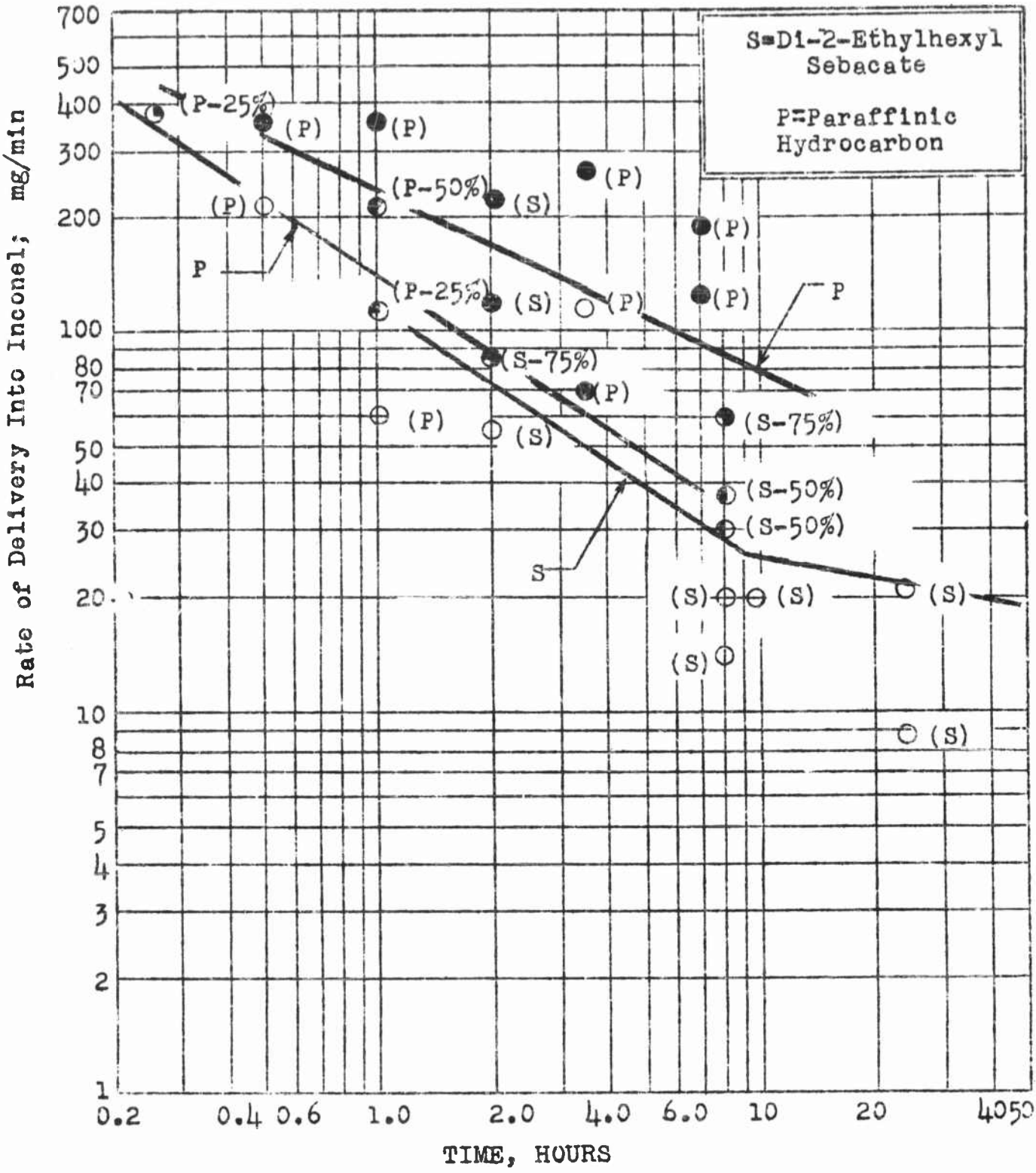


Figure 26. Fatalities Among Rabbits Following the Inhalation of the Fog Formed by Dropping Either a Paraffinic Hydrocarbon or Di-2-Ethylhexyl Sebacate Into an Inconel Tube Heated to 800° or to 700°F, Respectively

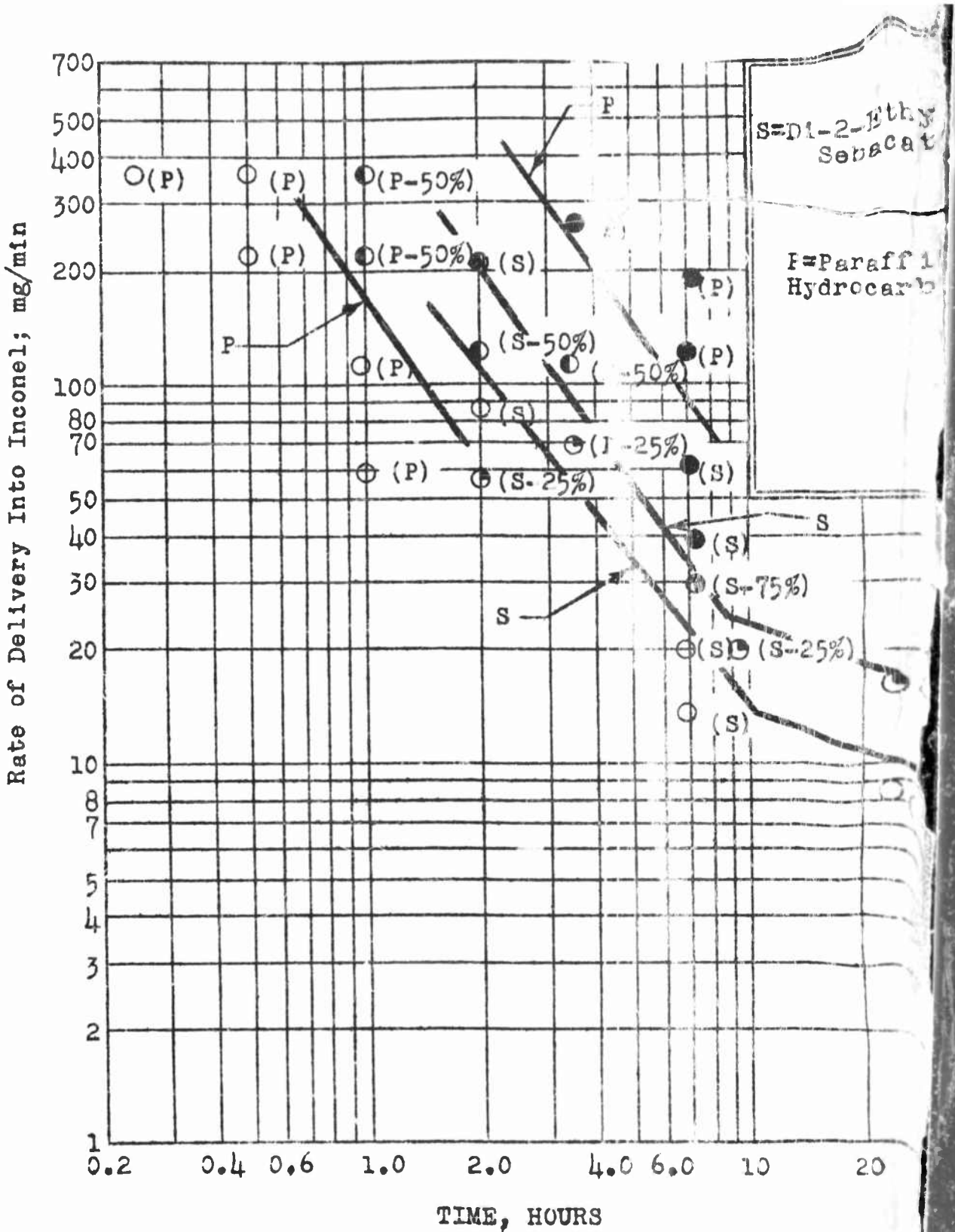


Figure 27. Fatalities Among Rats Following the Inhalation of the F
by Dropping Either a Paraffinic Hydrocarbon or Di-2-Eth
Sebacate Into an Inconel Tube Heated to 800° or to 700°
tively